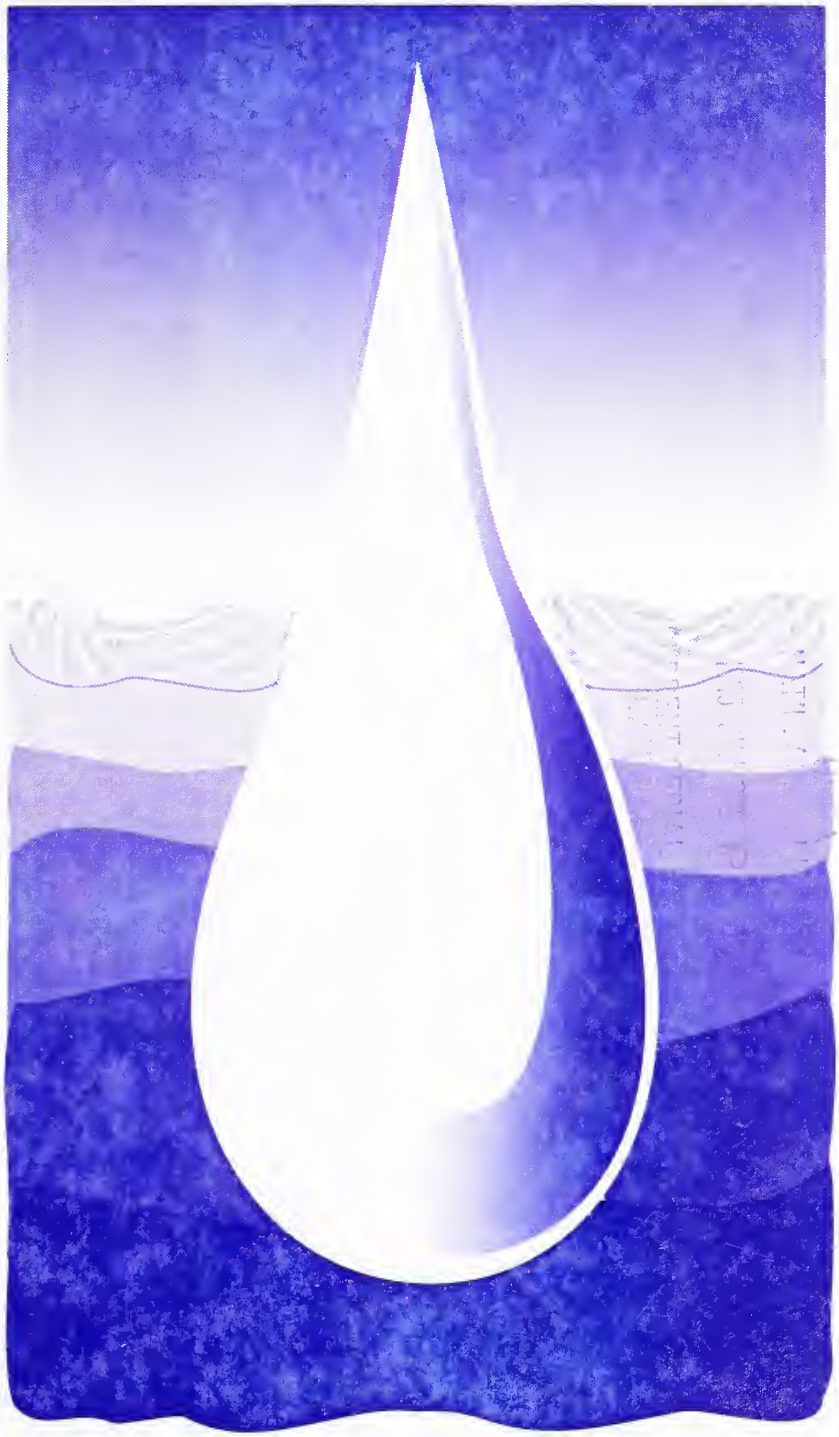
Natural Resources Institute

Water Quality Research



1991 Status Report

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U.S. Department of Agriculture
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Beltsville Agricultural Research Center

Natural Resources Institute

Water Quality Research

Status Report - 1991

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INTRODUCTION

This report covers the activities of scientists in the Natural Resources Institute, Beltsville Agricultural Research Center, who are involved in research related to water quality. The report has been divided into four (4) sections:

Section 1 covers the research accomplished in CY 1991. A short narrative of the purpose, accomplishment and application of the different research programs is is given. These narratives have been adapted from the 1991 CRIS Annual Reports.

Section 2 contains a list of papers published or accepted for publication in 1990-1991.

Section 3 contains a list of papers with interpretive summaries and technical abstracts that have been reviewed and approved for publication by ARS. These summaries are available in the ARS TEKTRAN (Technology Transfer Automated Retrieval System).

A search for "groundwater" papers in TEKTRAN listed 113 papers by ARS scientists for the period between January 1990 and January 1992, with 20 (18%) authored or co-authored by BARC scientists. However, I found 44 papers on groundwater by BARC scientists that have been approved and are available through the TEKTRAN system for the 2-year period.

Section 4 contains a list of scientists and their addresses and phone numbers.

SUMMARY

BARC scientists are actively involved in research to help better understand and manage water quality. Research is underway to 1) develop data bases, 2) develop expert systems, 3) develop and validate models, 4) measure and understand N and pesticide transport, 5) measure and understand volatilization and its impact on chemical loss, 6) develop methods to biodegrade pesticide wastes, 7) develop management systems to minimize N and pesticides loss from agricultural fields, 7) studies of biological effects, and 9) develop basic and theoretical understanding of chemical transport in agricultural, riparian, and natural ecosystems.

DISCLAIMER

Hopefully, this compilation is representative of the research on water quality at BARC. I have probably missed some that should have been included. If I have, let me know so that we can all know the complete water quality program at BARC. If you have any general questions, call me, Jerry Ritchie, on 301-5094-7490. If you have technical questions, call the scientist doing the research and see how you can become involved.

PREFACE

This is the second Status Report developed on the combined water quality research being conducted in several laboratories of the Natural Resources Institute (NRI), Beltsville Agricultural Research Center. Several important developments have occurred since the first report.

First, the magnitude of the water quality program is so large that a mutual decision was made between Federal and State personnel to combine research efforts. A Mid-Atlantic Consortium on Water Quality was established with the Natural Resources Institute (ARS), the Maryland Institute of Agriculture and Natural Resources (University of Maryland), Region III and the Office of Research and Development, U.S. Environmental Protection Agency (EPA), and the Mid-Atlantic Area, U.S. Geological Survey (USGS). Planning is now underway on joint regional projects.

Second, the theme of the Beltsville Symposium XVII will be Agricultural Water Quality Priorities, A Team Approach to Conserving Natural Resources. The symposium will be held May 4-8, 1992, in the newly renovated auditorium of Building 003, at the Beltsville Agricultural Research Center. During the symposium, over 100 oral and poster presentations will be made by leading scientists in the area of water quality.

The NRI water quality program is diverse in nature and covers a wide range of scientific research. New advances in NMR imaging of chemically induced tumor formation in fishy, the first isolation and description of genes encoding microbial enzymes responsible for atrazine degradation, new insights on the effects different tillage systems on pesticide movement, and better understanding of the effects of cover crops on nitrogen movement are a few of the features of the 1991 Status Report.

Finally, I wish to thank the National Program Staff of ARS for their support and encouragement of our water quality research programs. Specifically, I wish to identify the efforts of Drs. Dale Bucks, Doral Kemper, David Farrell and Dennis Childs of Dr. Jan van Schilfgaarde's Natural Resources and Systems Staff for providing increased financial support. The support and encouragement of Dr. Essex E. Finney, Jr., Area Director, and his staff has also been very helpful.

Dr. P. C. Kearney
Deputy Area Director
Natural Resources Institute

SECTION 1

RESEARCH ACCOMPLISHMENTS 1991

ARS PESTICIDE PROPERTIES DATABASE (ARS-PPD)

Pesticides are increasingly being found in trace amounts in our water supplies. To eliminate or reduce the risk of future water pollution, a database was developed that contained data on 16 properties of 92 pesticides. These properties are important to modelers and managers trying to predict the potential of various pesticides to move to the groundwater under a range of weather and soil conditions. The database has been released to SCS for use in management programs. Additional pesticides are being added to the data base. Data from the SCS/ARS/CES Pesticide Properties Database (Tifton, GA - Dr. Wauchope) are being incorporated into ARS-PPD with the intent of forming one database. Henceforth, all new data will be added solely to the ARS-PPD, which will continue to be administered in Beltsville.

CRIS 0500-00026-003-00 D Drs. Herner/Acock

NUTRIENT MANAGEMENT EXPERT SYSTEM (NUMEX)

Contamination of ground-water supplies by the application of fertilizers and pesticides at the farm level is a major concern. Nutrient Management Expert System (NUMEX), which is a combined laboratory information system and expert system, was developed to aid extension specialists in their recommendations to farmers on the application of commercial fertilizers, manure, and sewage sludge. NUMEX accumulates data on the residual fertilization content of each field soil submitted for analysis, the nutrient content of any manure or sewage sludge available to the farmers, and the history of the field. The expert system then makes recommendations to the farmers about the amounts of manure, sludge, and commercial fertilizer to apply in order to grow their crops without contaminating surface or ground water. NUMEX takes into account the slope and erodibility of the field and the leaching potential of the soil. It calculates the likely nitrate content of the soil based on field history and accumulation of heavy metals from sludge applications. The use of NUMEX at the farm level helps to reduce both the costs of chemicals and the amount of ground water pollution. Extension specialists from the University of Maryland provided the expert knowledge for NUMEX and the program is being used in Maryland for extension recommendations to farmers. The program is also being adapted to be used in Midwestern states.

CRIS 1270-61000-005-00 D Drs. Acock/Reddy

SOILSIM

Currently, the best crop simulation models have very simple representations of soil processes, and the best soil models have very rudimentary plants. As a result of these weaknesses, both types of these models suffer in their predictive capabilities. The Systems Research Laboratory scientists are trying to couple process-level crop simulation models with the soil model SOILSIM. The resulting union would improve crop models, but the real reward is as a better model of the movement of agricultural chemicals through soil and into water supplies. The soybean model GLYCIM and the cotton production model (CPM) are interfaced with SOILSIM.

CRIS 1270-61000-005-00 D Drs. Acock/Reddy

COVER CROPS CAN IMPROVE GROUNDWATER QUALITY

Autumn residual fertilizer nitrogen (FN) is vulnerable to leaching loss in humid climates and represents a wasted resource and a potential groundwater pollutant. Cover crops were evaluated for their ability to conserve residual FN. Average percent recovery of fall residual FN was 45% for cereal rye, 27% for annual ryegrass, 8% for hairy vetch, 7% for crimson clover, and 5% for native weeds (chickweed). These results confirm that grass cover crops, especially rye, are superior to legumes in conserving residual N. Grass cover crops can reduce both the mass of N leached and the nitrate concentration of the leachate by 20-80% compared to a no cover crop control. The EPIC model was used to simulate the impact of cover crops on nitrate leaching across the U.S. The model predicted that the greatest benefit of reduced nitrate leaching would occur in the Southeast and in irrigated agriculture. This demonstration of N conservation by grass cover crops should be useful to SCS, ASCS, and the Extension Service as they develop modern cropping systems for humid climates.

CRIS 1270-12130-004-00 D Dr. Meisinger

VARIATION IN DENITRIFICATION IN RIPARIAN ZONES

Denitrification within riparian zones may be a major and inexpensive nitrate pollution control measure available to land managers. A new method for on-site assessment of riparian denitrification rates has been developed, successfully tested in the laboratory, and samples from a December, 1991 field experiment are being analyzed. The method involves injection of acetylene into groundwater to block denitrification at the readily detectable nitrous oxide stage. Primary advantages of this experimental method are that it provides a way to assess in situ denitrification activity, and for repeated measurements at a particular site under different seasonal conditions

CRIS 1270-13000-003-00 D Drs. Starr/Bragan

EFFECTS OF TILLAGE ON PESTICIDE VOLATILIZATION

No-tillage (NT) practices are being implemented by farmers in an effort to save energy and time, and reduce soil erosion losses. The most noticeable characteristic of a NT field in comparison to a conventionally tilled (CT) field is the plant debris layer left on the soil surface. A side-by-side field experiment compared the volatilization losses of three pesticides from a CT plot and a NT plot. The plant debris layer on the NT plot intercepted 41-44% of the pesticide spray application. Volatilization losses were greater from the NT plot, especially for the more volatile pesticides. Larger volatilization losses from the NT plot appear to be due to the higher volatility of the fraction adsorbed to the surface plant debris than that adsorbed in the surface soil of the CT plot. These results suggest the need to use less volatile pesticides or different application techniques or pesticide formulations on NT fields.

CRIS 1270-13660-005-00 D — Ms. Schomburg

BEHAVIOR OF STARCH ENCAPSULATED ATRAZINE AND ALACHLOR

Public concern over the safety of drinking water supplies has resulted in intensification of the research effort directed at the development of formulations which modify the behavior of agricultural chemicals. Starch encapsulation (SE) is one such modification. A series of experiments have been conducted to determine the effect of temperature, water availability, and soil microbial activity on rate of release of herbicides from SE formulation and to compare volatilization of atrazine and alachlor, applied to the surface of moist soil as either emulsified concentrate (EC) or SE, at several temperatures. Rate of release of atrazine and alachlor from SE formulations increased as temperature, water availability, and microbial activity increased. Volatilization of atrazine and alachlor increased as temperature increased when applied as either EC or SE. Volatilization of atrazine was reduced when applied as SE compared to EC. Volatilization of alachlor was greater when applied as SE compared to EC. Effect of SE on herbicide behavior is dependent on the properties of the chemical being encapsulated.

CRIS 0500-00032-004-00 D — Drs. Wienhold/Gish

PRETREATMENT FOR QUANTITATIVE RECOVERY OF STARCH ENCAPSULATED HERBICIDES FROM SOIL

Starch encapsulation (SE) of herbicides is one formulation modification currently receiving significant research attention. Traditional extraction methods yield poor recovery of SE herbicides from soil. A laboratory study was conducted to develop a method for the quantitative recovery of herbicides applied to soil in the SE formulation. The pretreatment involves addition of the enzyme amylase in a phosphate buffer solution to the soil sample and incubation at 50^{o}C for one hour, followed by extraction with methanol water. Inclusion of the amylase pretreatment in the extraction procedure results in quantitative recovery of atrazine applied to soil as the SE formulation with a modest increase in time or cost of extraction.

CRIS 0500-00032-004-00 D — Drs. Wienhold/Gish

PREFERENTIAL MOVEMENT OF AGRICULTURAL CHEMICALS

Preferential transport encompasses a number of flow processes, each utilizing only a small fraction of the available pore space for chemical movement. A cooperative research study with the Pesticide Degradation Laboratory showed that under certain conditions, <1% of the soil volume was used to transport >9% of the surface spray applied herbicides. Although most of the herbicide remains in the surface 20 cm, the 9% alluded to was transported to a depth >1m in less than 6 days. Geostatistical analysis demonstrated that no-till treatment sites were more susceptible to preferential flow and transport of herbicides. Additionally, the long term study has shown that for pesticides the greatest potential for movement is in the spring, immediately after application, and in the winter during winter recharge.

CRIS 1270-13000-005-00 D Dr. Gish

FEASIBILITY OF USING STARCH ENCAPSULATED HERBICIDES TO MINIMIZE LEACHING

Due to the frequent observation of atrazine in groundwater, laboratory studies were conducted to evaluate the feasibility of using starch encapsulated atrazine to minimize leaching losses. Fifty surface soil cores were removed from an established no-till field site and used to evaluate leaching losses from three starch-encapsulated atrazine formulations relative to technical grade. Cumulative losses showed that after 16 pore volumes, 35% of the technical grade atrazine had leached through the shallow surface cores while between 1-3% for the jet-cooked, starch encapsulated process. Atrazine residue in the soil cores indicated that atrazine mobility was dramatically reduced by minimizing the amount available for convective transport, and maximizing adsorption affinity.

CRIS 1270-13000-005-00 D Dr. Gish

FIELD-SCALE IMPACT OF STARCH ENCAPSULATED HERBICIDES

In laboratory investigations starch encapsulated atrazine was found to minimize leaching losses in soil columns. However, nothing was known concerning the behavior of starch encapsulated atrazine and alachlor under non-isothermal, transient flow conditions. Recent field data from this experiment support laboratory results, i.e. convective transport of atrazine is greatly reduce compared to the common commercial formulation. No significant differences in weed control have been observed. However, atrazine persistence has increased as the pathways for atrazine loss have been reduced.

CRIS 1270-13000-005-00 D Dr. Gish

MEASUREMENT AND MODELING OF PESTICIDES IN THE FIELD

Model validation and testing with data from a long-term field study of pesticide transport under no-till (NT) and conventional till (CT) systems are continued. Our simulation results with PRZM compared quite well for the upper 10 cm but significantly underestimated atrazine residues in the lower depths of the root zones, especially in the NT plots. The reason for the differences in the model predictions for the lower depths is primarily due to the fact that PRZM does not account for rapid macropore fluxes. Furthermore, these model predictions of atrazine for the period of 1987-1989 were based on the estimated values of runoff and evapotranspiration, the two major components of the water balance in the PRZM model. Since 1990, additional equipment was added, for monitoring and sampling the runoff and several modifications were also made in the type and frequency of the data collections.
CRIS 1270-12130-003-00 D Drs. Isensee/Sadeghi

EFFECT OF TILLAGE PRACTICE ON RUNOFF PATTERNS AND HERBICIDE LOSS

Impact of tillage practice on runoff volume and pesticide loss was measured. Runoff from no-till (NT) and conventional-till (CT) plots was measured and atrazine, cyanazine, and alachlor concentrations in water were determined. Runoff from CT plots was 1.2 to >10 times greater than from NT plots when time since last rain was >7 days; runoff was 1.1 to 2 times greater from NT than CT plots when time was <7 days. Pesticide loss was highest for first runoff event after application. Atrazine and cyanazine concentrations were 2 to 5 times higher in runoff from NT than CT, while alachlor concentrations were about equal between tillages. Results should be useful in calibrating models that predict tillage practice effects on runoff and pesticide loss.
CRIS 1270-12310-003-00 D Drs. Isensee/Sadeghi

APPARATUS FOR STUDYING PESTICIDE LEACHING IN INTACT SOIL CORES

An apparatus was designed and constructed to study pesticide leaching through intact soil cores for the purpose of evaluating pesticide-soil-rainfall interactions as related to tillage practice. A turntable is used to support and rotate 8 to 12 soil cores under an oscillating rain simulator. Techniques were developed to mount the cores and collect leachate. Performance studies indicated good control of and low variability in rainfall rates and patterns. More atrazine was leached through conventional-till as compared to no-till cores by 1.2 pore volumes of water. This apparatus is a useful tool to study the effect of tillage practice on pesticide leaching because rainfall can be precisely simulated, a large number of cores can be simultaneously treated and it can be easily modified to meet research parameters.
CRIS 0500-00032-004-00 D Drs. Isensee/Sadeghi

PESTICIDE WASTE DISPOSAL PROCESS IMPROVED

Pesticide rinsate wastes have traditionally been discharged onto the soil to degrade or evaporate. This practice has frequently resulted in the contamination of nearby wells and soil surrounding the mixer-loader sites. Microbial degradation has historically been used as an effective disposal method for inherently biodegradable compounds, but this approach has not been effective for such persistent pesticides as atrazine. A two-step disposal process for pesticide waste was developed. It consisted of oxidation with ozone followed by microbial conversion of the material to its elemental forms. An important feature of the newly developed process include the use of an organism that degrades the ozonation products of atrazine in the presence of high levels of ammonia fertilizers, which ordinarily inhibit most microorganisms. Two new bioreacter designs with defined microbial supports were developed in the laboratory and are being scaled up for field testing as a means of disposing of atrazine and other persistent pesticides.

CRIS 1270-12130-003-00 D Drs. Hapeman-Somich/Shelton

ISOLATION OF A NEW BACTERIAL s-TRIAZINE DEGRADATION GENE

The widespread use and relative persistence of s-triazine herbicides such as atrazine has lead to increasing concern about surface and groundwater contamination by these compounds. Unfortunately, biodegradation studies have yielded very few microbial isolates that transform s-triazines. However, one bacterium, Rhodococcus corallinus, has been shown to have enzymatic activity that catalyzes the dechlorination of the s-triazine deethylsimazine (CEAT). We purified the CEAT dechlorination enzyme from extracts of R. corallinus and used information from the protein sequence to generate a DNA probe for the dechlorination gene. A large DNA fragment containing the dechlorination gene was isolated by using the DNA probe to screen recombinant bacteria containing Rhodococcus DNA fragments. DNA sequencing and expression analysis are now being used to characterize the dechlorination gene. Since dechlorination is frequently the rate limiting step in pesticide biodegradation of pesticides, the isolation and subsequent manipulation of this gene will provide us with a new biological tool to degrade s-triazine wastes.

CRIS 1270-12130-005-00 D Drs. Mulbry/Seffens

ERRATUM

In the process of printing this report, the unit for carbon dioxide concentration was inadvertently misprinted in a number of cases. The correct unit should be micromoles per mole (μmol mol^{-1}). This unit is equivalent to microliters per liter (μl l^{-1}), microbars per bar (μbar bar^{-1}), or part per million (ppm).

S-TRIAZINE DEGRADATION: GENE CLONING AND ENZYME CHARACTERIZATION

Triazine herbicides such as atrazine and simazine are used extensively in the culture of food crops. These compounds are slowly degraded by soil microorganisms, ultimately yielding carbon dioxide and ammonia. Cleavage of the s-triazine ring structure (a six-membered aromatic ring composed of alternating carbon and nitrogen) is an essential step for the complete mineralization of these herbicides. The enzyme cyanuric acid amidohydrolase cleaves this ring by a hydrolytic mechanism. The gene encoding this enzyme in a Pseudomonas bacterium was cloned into E. coli and over-expressed. The enzyme was purified and characterized. The enzyme was very specific for the s-triazine ring as it was unable to hydrolyze other related compounds. Understanding the biochemical basis of each step in the degradation of triazine herbicides may lead to the development of organisms and/or processes by which the rates of degradation can be controlled for maximum herbicidal action and protection of ground and surface waters.

CRIS 1270-12130-005-00 D Dr. Karns

JUDGING SUCCESSFUL N_2-FIXATION SYMBIOSES IN SOYBEANS EMPLOYING FOLIAR PHOTOSYNTHATE METABOLISM

A problem of world-wide magnitude has been the overuse of nitrate and ammonia fertilizers. In many parts of the world, nitrate contaminate both ground and surface water supplies. Alternatives are sought to limit the use of nitrogen fertilizers. Leguminous plants such as soybeans are able to unite symbiotically with N_2-fixing bacteria, and if the symbiosis is effective, the bacteria provide most or all of the ammonia needed by the plant, and there is no need to use nitrogen fertilizers. However, conditions in the farm field sometimes preclude an effective symbiosis between the host plant soybean, and its root nodule symbiont, Bradyrhizobium japonicum. A way is needed to judge the effectiveness of the symbiosis at an early stage of crop growth. We have found that monitoring the symbiont host soybean leaf starch and soluble protein accumulation, at 20-30 days post-emergence, appears to provide an early indicator of the success of the symbiosis. Relative to young (20-30 days old), non-N_2-fixing soybean plants supplied with growth sufficient nitrate and ammonia (N) levels, N_2-fixing plants with poorly established symbioses displayed very high foliar starch and very low protein levels. In contrast, a fully efficient symbiosis for young N_2-fixing soybean plants was accompanied by reduced leaf starch and elevated soluble protein levels similar to those of non-N_2-fixing, N supplied control plants. The work is leading toward the ability of legume crop breeders, soil microbiologists, soil scientists, extension specialists, and growers to assess both the early success of legume crop plant N_2-fixation symbioses , as well as the N fertilizer needs of the crop, by simplified monitoring of photosynthates.

CRIS 1270-21000-014-00 D Dr. Robinson

MAGNETIC RESONANCE IMAGING OF FISH SENTINELS

We have developed a procedure for the detection of liver tumors in live fish using magnetic resonance technology. This procedure used as a monitoring tool offers compatibility with other monitoring methods and studies that question the chemical, physical and biological effects of toxic substances. We are using magnetic resonance imaging (MRI) to correlate fish live anomalies with the presence of pollutants and contaminants in marine estuaries. We have correlated in vivo MRI of mummichog liver tumors with histological analyses. More recently, the initial analysis of a set of MRI parameters called relaxation times from fixed liver shows that data from classical histological procedures, which require killing the fish, and the MRI relaxation time data agree. Our project addresses the need for improved monitoring techniques and the need to gain insights into ecosystem health. These goals can be implemented by causal analysis that uses non-invasive, non-destructive methods. The bioindicator organism remains alive and unharmed following the experiment.

CRIS 1270-13660-005-00 D Drs. Gassner/Wright

EVALUATION OF THE POTENTIAL MUTAGENICITY OF GROUND AND SURFACE WATERS

The SOS microplate assay was used to determine the potential mutagenicity of ground and surface waters samples from well-defined agricultural areas. A protocol was established and standardized for the solid phase extraction of organic residues in water samples from: 1) BARC South Farm wells (samples monthly from March to August, 1991); 2) Northeast Watershed Research Center (NWRC) wells, Klingerstown, PA (sampled in November 1991); and 3) USGS wells on the Delmarva peninsula (Locust Grove and Williards, MD and Vandyke, DL; sampled in January, June, and October 1991). The relative activity (RA) of extracts as determined by the SOS microplate assay was expressed relative to the standard mutagen, 4-nitroquinoline oxide and a level of 0.04 or less was considered as inactive. Additionally organic carbon content was determined on the water samples. For 1991, the average range of RA for USGS wells was 0.04-0.065. The South Farm and NWRC wells were inactive. No major differences in RA were observed between South Farm wells in Conventional Till (CT) and No till (NT) plots except for a spike of RA in South Farm wells in CT plots in August 1990. A similar spike of RA was not found in 1991. Spatial and temporal variations in the RA were observed at all sites and there appears to be no correlation between carbon content and RA.

CRIS 1270-13660-006-00 D Drs. Pfeil/Nair

SECTION 2

PUBLICATIONS 1990-1991

BARC PUBLICATIONS ON WATER QUALITY 1990-1992

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SECTION 3

ARS APPROVED PUBLICATIONS

1990-1991

SECTION 3

ARS APPROVED PUBLICATIONS

1990-1991

Agricultural Research Service
Technology Transfer Automated Retrieval System - (TEKTRAN)

THE VALUE OF LONG-TERM FIELD RESEARCH IN MODEL DEVELOPMENT AND VALIDATION

ACOCK BASIL

Interpretive Summary:
N/A
Technical Abstract:
When many long-term field experiments were initiated, the principal method of analyzing the data was to use multiple regression analysis. In this technique it assumed that the interaction between environmental factors can be described with empirical additive or multiplicative models. In the early days of agricultural experimentation, when gross nutrient deficiencies were being corrected, the exact form of the model did not matter much. Now, with nutrient levels near optimum, additional factors have to be considered and our models must deal with the interaction between all factors realistically. The most realistic way of considering interactions between a large number of factors is to use a limiting factor model. This enables us to develop mechanistic models which simulate many of the processes going on in the plant, soil, and atmosphere. Field data are essential to the final stages of model development and validation. Long-term field research is especially useful because it provides: 1. Data in which some environmental factors are constant, and observed effects can be attributed to uncontrolled factors, and 2. Data from which the unknown effects of previous treatments have been eliminated. Modelers would benefit from working more closely with scientists running long-term field experiments: showing them what additional data are needed and how existing models can be used to interpret results.

Submitted to: (approved 03/01/90)
AGRONOMY JOURNAL (SPECIAL EDITION)
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USDA:ARS:BA:NRI:SRL
BLDG 011A,RM 165-B,BARC-W FAX Number:
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Agricultural Research Service
Technology Transfer Automated Retrieval System - (TEKTRAN)

STRUCTURING AGRICULTURAL RESEARCH TO DELIVER DECISION-SUPPORT PRODUCTS

ACOCK BASIL

Interpretive Summary:

Technical Abstract:
Agricultural research supports farmers by providing them with a rational basis for management decisions. Judging by our food surpluses, the present system for delivering research results to the farmer is highly successful. That success has come from solving yesterday's problems, but it has left us with other, more intractable, problems to solve today. The most serious of these is contamination of our environment with toxic and carcinogenic chemicals. We can also foresee the need to prepare for future global climate change and for managing genetically engineered organisms. The present decision-support products and the system for delivering them are inadequate to meet these challenges, but new tools are becoming available. Rapid advances in computer power and availability have given us the opportunity to manage complex operations like farms in a way unimaginable only ten years ago. Since all processes can be described in mathematical terms, computers can be used to mimic the behavior of plants and animals and to help farmers decide between possible management options. In this chapter I analyses the farm decision-support system and suggest improvements that can be made by treating crops, animals, etc. as sub-systems, and by using systems analysis and simulation to better understand and manage them.

Submitted to: (approved 03/01/90)
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Agricultural Research Service
Technology Transfer Automated Retrieval System - (TEKTRAN)

CUG AS A MUTANT START CODON FOR CAT-86 AND XYLE IN BACILLUS SUBTILIS

AMBULOS NICHOLAS JR
SMITH TIMOTHY
MULBRY WALTER
LOVETT PAUL S

Interpretive Summary:
The biochemical mechanisms, which underlie the genetic processes of bacteria and more complex organisms, are the focus of intense study worldwide. Within this realm of research are studies of the processes by which the genetic information encoded on the DNA of an organism is deciphered so that such cellular material as proteins are made. Recombinant DNA techniques were used in this study to demonstrate that the 'genetic punctuation' used to mark the location on DNA that protein synthesis should start varies between two different important groups of bacteria. This study revealed that a DNA sequence, which does not function as a genetic punctuation signal in many such organisms as the well studied intestinal bacteria Escherichia coli, works well as a signal in the industrially important bacteria belonging to the group Bacillus. This finding is important to scientists who examine DNA while searching for important genes and to those who are trying to manipulate important genes so that they may be expressed in useful organisms

Technical Abstract:
The cat-86 gene specifies chloramphenicol acetyltransferase (CAT). The cat-86 start codon is UUG, although related genes have AUG as the start codon. Changing the start codon to GUG and CUG increased expression of cat-86 by 25% in Bacillus subtilis. Changing the start codon to GUG and CUG decreased expression to 65% and 30%, respectively, of the level obtained when AUG was the start codon. CUG has not been previously shown to function as a start codon in B. subtilis. N-terminal sequencing of purified CAT protein specified by the CUG mutant, revealed that CUG was indeed the start codon and specified methionine. The gene xylE, which specifies catechol 2,3,-dioxygenase, has AUG as its start codon. Changing the start codon for xylE to CUG decreased expression by 98%. However, when the ribosome binding site sequence of xylE was optimized and the spacing between it and the start codon was increased to 8 nucleotides, xylE activity increased to 13% of the activity observed for AUG. CUG did not function efficiently as a start codon for cat-86 in Escherichia coli. These data suggest conditions under which CUG can function, with modest efficiency, as a start codon in B. subtilis.

Submitted to: (approved 12/27/90)
GENE

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Technology Transfer Automated Retrieval System - (TEKTRAN)

BASIC SOIL FERTILITY PRINCIPLES

BANDEL VERNON A
JAMES BRUCE R
MEISINGER JOHN J

Interpretive Summary:
Farmers need to know about basic soil fertility in order to manage nutrients for farm profitability and to maintain the quality of our environment. This extension service bulletin reviews the sources of nutrients, transformation and fate of nutrients, and the plant availability of the 13 essential nutrients. These 13 nutrients consist of the primary nutrients nitrogen, phosphorus and potassium; the secondary nutrients of calcium, magnesium and sulfur; and the micro nutrients of copper, zinc, manganese, boron, iron, molybdenum, and chloride. The importance of soil pH, soil organic matter, soil cation exchange capacity, and soil texture are also discussed. The plant availability of these nutrients is assessed through soil tests which are the best way to inventory a farms' soil resources on a regular basis. Understanding basic soil fertility will help farmers improve nutrient use efficiencies in Maryland and should reduce nutrient losses to Chesapeake Bay.

Technical Abstract:
Managing soil fertility is essential for profitable agriculture and to maintain our environment. This bulletin reviews basic soil fertility principles for the 3 primary nutrients (N, P and K), the 3 secondary nutrients (Ca, Mg and S) and the 7 micronutrients (Cu, Mn, Zn, B, Fe, Mo and Cl) including their transformations in soil and their plant availability as related to soil pH, soil organic matter, soil texture, soil cation exchange capacity (CEC) and soil aeration. The soil testing procedures for these 13 nutrients are reviewed, as well as the testing procedures for pH, organic matter, CEC, and texture. Soil tests must be calibrated against actual crop response in the field before a fertilizer recommendation can be made. It is important for modern farmers to inventory their soil resources through regular soil testing and to wisely use manures and fertilizers as nutrient resources. This publication will help train farmers and extension service personnel to improve nutrient use efficiencies in Maryland and should reduce nutrient losses to the environment.

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GENETIC ENGINEERING APPROACH TO TOXIC WASTE MANAGEMENT: CASE STUDY FOR ORGANOPHOSPHATE WASTE TREATMENT

COPPELLA STEVEN J
DELACRUZ NESLIHAN
PAYNE, GREGORY F
POGELL BURTON M
SPEEDIE MARILYN K
KARNS JEFFREY S
SYBERT EDWARD M
CONNOR MICHAEL A

Interpretive Summary:
Genetic engineering may offer the means for developing methods for the rapid and convenient disposal of excess or waste agricultural chemicals. A method was developed whereby the enzyme parathion hydrolase was used to aid in the degradation of waste coumaphos, an acaricide used to protect cattle from ticks. The gene encoding parathion hydrolase was cloned from its native Flavobacterium and placed into Streptomyces lividans. The protein encoded by the was overproduced in recombinant construct and was excreted into the culture medium. Both properties are useful for the production of enzymes on an industrial scale. The conditions for culture of the recombinant organisms were adjusted to obtain maximal production of parathion hydrolase. The cell-free culture fluid from the fermentation of recombinant microorganism was effective in hydrolyzing waste coumaphos. By treating the waste with an enzyme rather than the whole organism the problems associated with the release of recombinant microorganisms into the environment are avoided.

Technical Abstract:
Currently, there has been limited use of genetic engineering for waste treatment. In this work, we are developing a procedure for the in situ treatment of toxic organophosphate wastes using the enzyme parathion hydrolase. Since this strategy is based on the use of an enzyme and not viable microorganisms, recombinant DNA technology could be used without the problems associated with releasing genetically altered microorganisms into the environment. The gene coding for parathion hydrolase was cloned into a Streptomyces lividans, and this transformed bacterium was observed to express and excrete this enzyme. Subsequently, fermentation conditions were developed to enhance enzyme production, and this fermentation was scaled-up to the pilot scale. The cell-free culture fluid (i.e., a nonpurified enzyme solution) was observed to be capable of effectively hydrolyzing organophosphate compounds under laboratory and simulated in situ conditions.

Submitted to: (approved 08/24/90)
BIOTECHNOLOGY PROGRESS

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Technology Transfer Automated Retrieval System - (TEKTRAN)

PHOTOSYNTHATE METABOLISM IN THE SOURCE LEAVES OF N2-FIXING SOYBEAN PLANTS

DE VEAU EDWARD J
WARMBRODT ROBERT D
ROBINSON J MICHAEL
KREMER DIANE F

Interpretive Summary:
A problem of national magnitude has been the extreme overuse of nitrate and ammonia fertilizers in the farms of North America. In many parts of the U.S., nitrates severely contaminate ground water supplies, and alternatives are sought to limit the use of nitrogen fertilizers. Leguminous plants such as soybean are able to unite symbiotically with N2-fixing bacteria. If the symbiosis is effective, the bacteria provide most or all of the ammonia needed by that plant, and there is no need to use nitrogen fertilizers. However, conditions in the farm field sometimes preclude an effective symbiosis between the host plant soybean, and its symbiont, root nodule, bacteria, Bradyrhizobium japonicum. A way to determine the effectiveness of the symbiosis by examination of the plant leaves is needed. This work reports that soybean leaf synthesis of carbohydrates such as starch is a measure of the effectiveness of the symbiotic relationship. The work is leading toward the ability of farmers and grower groups to assess the need to supply additional nitrate and ammonia fertilizer to the nitrogen-fixing soybean plant by monitoring the leaf carbohydrate status.

Technical Abstract:
Glycine max [L.] Merr. cv Williams plants, which were symbiotic with Bradyrhizobium japonicum, and which acquired reduced nitrogen solely through N2 fixation processes (N2Fx plants), often exhibited excess accumulation of starch and sucrose in their source leaves when compared with counterpart soybean plants which had been supplied from emergence with growth sufficient levels of inorganic nitrogen (6 mM NO3-/6 mM NH4+)(NS plants). Since there was a minimal requirement for carbon skeletons to support amino acid synthesis, enhanced foliar starch accumulation appeared to be related to a considerable excess of hexose phosphates generated during CO2 assimilation. Evidence suggested that increased starch accumulation in N2Fx plants was facilitated by: 1) an adaptive increase of activities of several enzymes of the chloroplast starch synthesis pathway including, fructose-1,6-bisphosphate(C-1) phosphatase, phosphohexoisomerase, phosphoglucomutase and adenosine diphosphate glucose pyrophosphorylase (ADPG-PPiase) (in some leaves), and 2) increased foliar levels of hexose monophosphates, e.g. fructose-6-phosphate, glucose-6-phosphate, and glucose-1-phosphate (G-1-P) which apparently had risen to levels considerably in excess of the Kms for their respective target enzymes, e.g. G-1-P with respect to ADPG-PPiase.

Submitted to: (approved 09/17/91)
PLANT PHYSIOLOGY

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Technology Transfer Automated Retrieval System - (TEKTRAN)

PHOTOSYNTHESIS AND PHOTOSYNTHATE PARTITIONING IN N2-FIXING SOYBEANS

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WARMBRODT ROBERT D
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Interpretive Summary:
Soybeans have two sources of inorganic nitrogen upon which to draw for vegetative growth as well as pod and bean development: 1) inorganic nitrogen (ammonia, nitrate, urea, etc.) in the soil and 2) symbiotic nitrogen fixation by the bacterium Bradyrhizobium in the root nodules. Less reliance on nitrogen fertilizers and more on the bacterium-soybean symbiosis to provide the nitrogen needs of soybeans would reduce production costs and result in the reduction of inorganic nitrogen reaching and polluting the waters in this country. It is not well known how the plant allocates photosynthate (photosynthetic products) to support the two different ways of acquiring nitrogen for growth. ARS scientists have found that, in the vegetative state, the soybean plant assimilation of inorganic nitrogen requires more photosynthate than the symbiotic fixation of atmospheric nitrogen. Soybean breeders can use this information to generate soybean varieties which can mobilize more of their photosynthate reserves for the support of nitrogen fixation and overall growth. Such efforts are expected to reduce production costs and nitrogen pollution of surface and ground waters.

Technical Abstract:
Leaf area, Chl content, net photosynthesis, and the partitioning of fixed carbon between foliar carbohydrate and protein were examined in Glycine max cv. Williams grown under three different nitrogen (N) regimes. One group (Nod+/+) was inoculated with Bradyrhizobium and watered daily with a nutrient solution containing 6 mM NH4NO3. A second set (Nod+/-) was inoculated and had N2 fixation as its sole source of N. A third group (Nod-) was not inoculated and was watered daily with a nutrient solution containing 6 mM NH4NO3. Net umol CO2 uptake/dm2.h of the most recently matured source leaves was similar among the three groups of plants, being about 310. Leaf area of the source leaves was also similar. However, the mg Chl/dm2 content of Nod+/- source leaves was 50% lower than the other treatments and indicated N deficiency. Thus, compared with Nod- and Nod+/+ plants, Nod+/- utilized less Chl to maintain a normal rate of photosynthetic CO2 uptake. The ratio of foliar carbohydrate:protein content was high in Nod+/- but low in the other two treatments. This suggests that more fixed carbon is diverted to the synthesis of protein when N availability is high. It is clear that both leaf carbohydrate and protein must be considered as end products of carbon assimilate partitioning when inorganic N assimilation is the controlling factor.

Submitted to:
PLANT PHYSIOLOGY
(approved 03/09/90)

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CLONING AND ANALYSIS OF S-TRIAZINE CATABOLIC GENES FROM PSEUDOMONAS NRRLB-12227

EATON RICHARD W KARNS JEFFREY S

Interpretive Summary:
The s-triazine herbicides atrazine, simazine, and cyanazine comprise a major portion of the herbicides used at the present time. These compounds have been shown to be very resistant to microbial degradation and few, if any, microbes have been shown to completely degrade these compounds. Swiss researchers isolated several organisms that can degrade the simpler s-triazine compounds that are likely to be intermediates in the biodegradation of s-triazine herbicides. The most versatile of these organisms is Pseudomonas sp. NRRLB-12227 which degrades the s-triazines melamine, ammeline, ammelide, and cyanuric acid through biuret to form urea. The organism uses the s-triazine compounds as a sole source of nitrogen but must have a carbon source supplied. In this paper we describe the cloning and characterization of several genes which encode enzymes involved in the degradation of ammeline, ammelide and cyanuric acid in Pseudomonas sp. NRRLB-12227. In addition the discovery of a repeated DNA element that apparently causes deletion and rearrangement of DNA in close proximity to the s-triazine degradation genes is described. The structure of the s-triazine genes flanked by this repeated DNA element physically resembles a transposon, which is a mobile DNA element. This research suggests that DNA recombination events play an important role in the evolution of new biodegradation pathways in bacteria. It is hoped that cloned s-triazine degradation genes might be used in the future to construct strains of bacteria capable of directly degrading s-triazine herbicides.

Technical Abstract:
Pseudomonas NRRLB-12227 degrades the s-triazine melamine, by a six step pathway which allows it to use melamine and pathway intermediates as nitrogen sources. Using the plasmid pLG221, mutants defective in five of the six steps of the pathway were generated. Tn5-containing-EcoR I fragments from these mutants were cloned and identified by selecting for Tn5-encoded kanamycin resistance in transformants. A restriction fragment from ammelide mutant RE411 was used as a probe in colony hybridization experiments to identify cloned wild-type s-triazine catabolic genes encoding ammeline aminohydrolase, ammelide aminohydrolase, and cyanuric acid amidohydrolase. These genes were cloned from total cellular DNA on several similar, but not identical, Hind III fragments, as well as on a Pst I fragment and a Bgl II fragment. Restriction mapping and Southern hybridization analyses of these cloned DNA fragments suggest that these s-triazine catabolic genes may be located on a transposable element, the ends of which are identical 2.2 kb insertion sequences.

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JOURNAL OF BACTERIOLOGY

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Technology Transfer Automated Retrieval System - (TEKTRAN)

CLONING AND COMPARISON OF THE DNA ENCODING AMMELIDE AMINOHYDROLASE AND CYANURIC ACID AMIDOHYDROLASE FROM THREE S-TRIAZINE-DEGRADING BACTERIAL STRAINS

EATON RICHARD W KARNS JEFFREY S

Interpretive Summary:
The s-triazine herbicides atrazine, simazine, and cyanazine comprise a major portion of the herbicides used at the present time. These compounds have been shown to be very resistant to microbial degradation and few, if any, microbes have been shown to completely degrade these compounds. Swiss researchers isolated several organisms that can degrade the simpler s-triazine compounds that are likely to be intermediates in the biodegradation of s-triazine herbicides. In this paper we show that three of these organisms (two distinct Pseudomonas species and a strain of Klebsiella pneumoniae) share identical genes for ammelide aminohydrolase and cyanuric acid amidohydrolase. In one of the Pseudomonas strains a gene for ammeline aminohydrolase has been inserted in the DNA adjacent to these other two genes. This research suggests that DNA recombination events play an important role in the evolution of new biodegradation pathways in bacteria and in the spread of these new pathways among the microbial flora of soil and water.

Technical Abstract:
DNA encoding the catabolism of the s-triazines, ammelide and cyanuric acid, was cloned from Pseudomonas NRRLB-12228 and Klebsiella pneumoniae 99 using as probe a 4.6 kb Pst I fragment from a third strain, Pseudomonas NRRLB-12227, which also encodes these activities. In strains NRRLB-12228 and 99 the ammelide aminohydrolase (trz C) and cyanuric acid amidohydrolase (trz D) genes are located on an identical 4.6 kb Pst I fragment which is part of a 12.4 kb DNA segment present in both strains. Strain NRRLB-12227 also carries this 12.4 kb DNA segment except that a DNA segment of 0.8 to 1.85 kb encoding a third enzyme, ammeline aminohydrolase (trz B), has been inserted next to the ammelide aminohydrolase gene with the accompanying deletion of 1.1 to 2.15 kb of DNA. In addition, the s-triazine catabolic genes are flanked in strain 12227 by identical 2.2 kb segments, not present in the other two strains, that appear to cause rearrangements in adjacent DNA.

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JOURNAL OF BACTERIOLOGY

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FEASIBILITY STUDY: MAGNETIC RESONANCE IMAGING OF HEPATIC NEOPLASMS IN MUMMICHOG FUNDULUS HETEROCLITUS FROM A CREOSOTE-CONTAMINATED ENVIRONMENT

GASSNER GEORGE
VOGELBEIN WOLFGANG K
LINE MICHAEL J
MILLARD MERLE M
VAN VELD PETER A
PLIMMER JACK R

Interpretive Summary:
Magnetic Resonance Imaging and histopathology were combined to detect and classify hepatic neoplasms and preneoplastic lesions in Elizabeth River mummichog. The work showed that the species can tolerate the physical manipulations required for MRI and determined that hepatic neoplasms can be detected in live, anesthetized fish and chemically-fixed whole fish livers.

Technical Abstract:
Adult mummichog, Fundulus heteroclitus inhabiting a creosote-contaminated site in the Elizabeth River, U.S.A. exhibit high prevalences of putative preneoplastic (73%) hepatic lesions. Sediments at this site are heavily contaminated with polycyclic aromatic hydrocarbon (2200 mg/kg dry sediment), several of which are potent chemical carcinogens. We are examining fixed tissues and live animals from this habitat in a Bruker 400 MSL microimaging spectrometer and correlating the resulting NMR images with histopathological diagnoses in order to: 1) determine if pre-neoplastic hepatic lesions (foci of cellular alteration) can be distinguished from hepatic neoplasms by this non-invasive technique, 2) evaluate the utility NMR imaging in studies of lesion progressing and 3) apply NMR imaging as an in vivo monitoring system environmental exposure to chemical carcinogens using a representative aquatic organism.

Submitted to: (approved 03/26/91)
PROCEEDINGS OF CHESAPEAKE RESEARCH CONSORTIUM BALTIMORE MD DEC 4-6 1990

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MAGNETIC RESONANCE DETECTION OF ENVIRONMENTALLY INDUCED HEPATIC LESIONS IN MUMMICHOG

GASSNER GEORGE
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Interpretive Summary:
Magnetic resonance imaging (MRI), a method for physical and chemical analysis of a living subject without violating its integrity, is being used to correlate fish liver anomalies with the presence of pollutants and contaminants in marine estuaries. The initial analysis of a set of MRI parameters called relaxation times shows that data from classical histological procedures, which require killing the fish, and the MRI data agree. These base line data sets establish a new generation of analyses for the safe use of agricultural chemicals in relation to water quality.

Technical Abstract:
A positive association between chronic exposure to polycyclic aromatic hydrocarbon (PAH)-contaminated sediments and the occurrence of hepatic neoplasms in a population of Elizabeth River mummichog was recently described by Vogelbein. High prevalences of putative preneoplastic (73%) and neoplastic (35%) hepatic lesions were observed in adult fish from a highly PAH-contaminated site adjacent to a wood treatment facility. Fish from two less contaminated sites did not exhibit these lesions. The present study combines histopathology and magnetic resonance images to diagnose and detect environmentally-induced hepatic lesions in mummichog livers. Both histologically fixed livers and in situ livers of living fish can be evaluated using MR techniques. Mummichog exhibit no detrimental effects after several months of observation following our in vivo MR procedure. The MR imaging data and histological observations on liver pathology correlate. This work establishes a non-invasive and non-destructive procedure for progressive and regressive studies of fish liver pathology and provides baseline information for the characterization and use of altered fish liver cellular architecture as an indication of PAH pollution in the marine environment.

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IMPACT OF PREFERENTIAL TRANSPORT OF PESTICIDES ON WATER QUALITY

GISH TIMOTHY J

Interpretive Summary:
A three-year cornfield study in Maryland showed that in dry years, pesticides levels in a shallow perched ground water were negligible, regardless of tillage practice. However, in 1988 a long-duration, low-intensity rain event occurred 12 hours after pesticide application, resulting in significant concentrations of all parasiticide in the perched water table. Additionally, pesticide levels were highest under no-till management; atrazine was four times higher under no-till than conventional tillage; cyanazine and alachlor almost three times higher; and carbofuran almost 13 times higher. The enhanced movement of the pesticides in 1988 was a result of preferential transport, a process whereby the chemicals move rapidly through void root channels, and soil cracks, bypassing much of the soil volume which has a high affinity for these compounds.

Technical Abstract:
A three-year field study was conducted to determine the impact of tillage practice, mode of pesticide application, and pesticide formulation of chemical transport. Pesticides evaluated were atrazine, cyanazine, carbofuran, and alachlor. Atrazine and cyanazine were applied as solutions whereas alachlor was applied as an emulsifiable concentrate in 1986, and as a microencapsulated formulation in 1987 and 1988. Carbofuran was band-injected at planting the day prior to the other three compounds. Drought conditions were experienced in 1986 and 1987; at which time only atrazine appeared in any significant concentrations, mean field-scale values ranging from 0.4 to 2 ug L -1. In May 1988, 12 hours after application, an extended, low-intensity, rain event occurred which administered 4.8 cm of water to the field site. Six days after the 1988 application, significant concentration of all pesticides were detected at approximately 1 m. Atrazine and cyanazine showed significant preferential transport, where concentration levels were almost an order of magnitude higher than those under conventional tillage. At least 9% of the available atrazine and cyanazine had leached to the shallow water table, utilizing <1% of the total soil vol. Incorporating carbofuran, and utilizing microencapsulated alachlor appears to have minimized the effect of pref. transport

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TRANSACTION OF AMER SOC OF AG ENGNRS

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MODELING PREFERENTIAL MOVEMENT OF AGRICULTURAL CHEMICALS

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HELLINGS CHARLES S

Interpretive Summary:
For the past decade there has been a great deal of public and political concern over the impact of agriculture on the environment. This concern has lead to a need for mathematical models having the ability to quantify agricultural chemical behavior in soil. However, the lack of adequate field data has made it impossible to quantify field-scale transport processes, so the assumptions inherent in laboratory studies have been extended to the field. Unfortunately, the fluid dynamics of a laboratory soil column are much different than a non-homogeneous field soil. Consequently, laboratory results have shown to be a poor indicator of field-scale movement of agricultural chemicals. Recent field-scale research is beginning to show which chemical transport processes are important on a large scale. This manuscript uses the observed results to determine a more realistic derivation of a field-scale chemical transport model.

Technical Abstract:
The classical convection-dispersion equation (CDE) has proven to be inadequate in simulating field-scale behavior. Part of the failure in field-scale models using the CDE is that detailed information about chemical behavior is sacrificed so as to account for other chemical and biological processes. Unfortunately, little is known concerning which process can be sacrificed without significantly effecting simulation results. Consequently, field-data from two recently conducted experiments was used to evaluate the classical CDE. Results indicate that simulation of field-scale chemical non-equilibrium, and non-homogenous flow regime may be drastically improved by using a two site mobile-immobile representation with two dissipation and adsorption parameters. The adsorption and dissipation coefficients are partitioned between the inter and intra-aggregate pore space.

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ASAE SYMPOSIUM PROCEEDINGS

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TRANSPORT COMPARISON OF STARCH-ENCAPSULATED ATRAZINE

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WING ROBERT E
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Interpretive Summary:
Atrazine one of the oldest and most frequently studied herbicides, is also one of the most common pesticide contaminates in ground water. Typically observed concentration levels are below EPA's health advisory level. However, recent field studies have demonstrated that the EPA levels could be substantially exceeded if used in an environment conducive to preferential flow. As a result, a laboratory study was conducted to test the feasibility of utilizing starch-encapsulated atrazine, to minimize leaching under environment conditions favoring preferential transport. Technical grade atrazine and three starch-encapsulated formulations were applied to 40 small soil cores, removed from an established no-till field site. Seven rain simulations were applied to these columns over a 21 day period, the leachate collected after each irrigation event. The greatest atrazine leaching took place during the first irrigation on the technical grade treatment, atrazine levels exceeding all encapsulation treatments by over a factor of 10. Over the duration of the experiment, encapsulating atrazine in a starch matrix, reduced leaching losses from 35% to < 1%. In the first irrigation, the atrazine in the leachate utilized < 10% of the soil volume supporting the hypothesis of preferential movement.

Technical Abstract:
The feasibility of using starch-encapsulated atrazine to minimize convective transport under conditions favoring preferential flow was evaluated. Forty small undisturbed cores were removed from an established no-till field site and randomly grouped into one of five treatment formulations: i) technical grade atrazine; ii) borate process, starch-encapsulated; iii) jet-cooked pearl starch-encapsulated; iv) jet-cooked, waxy starch-encapsulated; and v) untreated controls. Columns were irrigated with 0.01 N CaCl2 applied through a drip system, at the rate of 2.5 cm every three days Column effluent was collected and analyzed for atrazine as a function of time. Highest atrazine residue levels, 1.30 mg L-1, were observed in the effluent of the nonformulated herbicide after the first irrigation. Piston and convection-dispersion transport simulations predicted transit times that where almost an order of magnitude later than indicated from observed atrazine concentrations in the technical grade treatment. Starch-encapsul ation reduced atrazine formulations revealed little temporal variability in the effluent concentrations, while the technical grade demonstrated temporal coefficients of variation of 99%.

Submitted to: (approved 07/20/90)
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RELEASE AND PERSISTENCE COMPARISON OF STARCH-ENCAPSULATED ATRAZINE

GISH T J
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SCHREIBER M M
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Interpretive Summary:
With the increased interest in water quality issues, polymer chemists and soil scientists are combining their expertise to develop controlled release pesticide formulations. By encapsulating pesticide in a starch matrix, its release into the environment can be controlled. However, the release of pesticide from the starch matrix will be governed by a number of variables, such as: type of encapsulation process, type of starch, and soil physical properties. This investigation focuses on the affect of the water energy status on the release and persistence of several starch-encapsulated atrazine formulations. Results indicate that as the energy status of the water decreases so does the atrazine release rate. In addition, the persistence of the starch-encapsulated atrazine relative to non-encapsulated treatment appears to be a short term phenomena, due to the biological decomposition of the starch matrix.

Technical Abstract:
The release of atrazine as a function of water potential, and laboratory persistence rates were evaluated for three starch-encapsulated formulations. Capsule swelling and atrazine release rates from borate, jet-cooked pearl starch, and jet-cooked waxy starch were evaluated at several water potentials from 0 to -1.5 MPa. Generally, both the rate of atrazine release as well as the maximum equilibrium solution concentration decreased with decreasing water potential. Although starch-encapsulated atrazine is more persistent than non-encapsulated, it appears to be a short term phenomenon, related to the biodegradability of the starch matrix.

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WEED SCIENCE

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ATRAZINE RELEASE FROM STARCH-ENCAPSULATED FORMULATIONS AND PERSISTENCE IN SOIL

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HELLING CHARLES S
SCHREIBER MARVIN M
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WING ROBERT E

Interpretive Summary:
Previously, the rapid movement of atrazine was severely retarded by the use of starch-encapsulated formulations. However, the environmental fate of pesticides is a combination of mobility and persistence. Pesticide release and persistence in soil is a function of a number of soil and climatic factors. This study was conducted to evaluate the impact of several of these factors on the release and persistence of atrazine from newly developed ARS starch-encapsulated formulations. Results indicate that one of the new starch encapsulated process (jet-cooking) increases atrazine persistence in soil. As a result, field-scale interaction between surface and subsurface hydrology will be critical in determining the applicability of this formulations in agriculture.

Technical Abstract:
Atrazine release as a function of water potential, and atrazine persistence rates in soil, were evaluated for three starch-encapsulated formulations. Capsule swelling and atrazine release rates from borate, jet-cooked pearl, and jet cooked waxy formulations were evaluated at several water potentials from 0.0 to -1.5 MPa. Rate of atrazine release as well as the maximum equilibrium solution concentration of the herbicide decreased with decreasing water potential. Relative to published field observations, atrazine persistence in this laboratory study was lower due to high soil moisture conditions, high organic matter content, and slightly higher temperatures. Persistence rates were greatest for the jet-cooked encapsulated atrazine and shortest for technical grade. Enhanced atrazine persistence associated with encapsulation appears to be a short-term phenomenon, inversely related to the biodegradability of the starch matrix. Nomenclature: Atrazine, (6-chloro-N-ethyl-N'-(1-methylethyl)-1,3,5-triazine-2,4-diamine).

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WEED SCIENCE
(approved 07/10/91)

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PRELIMINARY STUDIES OF THE DISTRIBUTION, DRIFT, AND VOLATILIZATION OF DIAZINON RESULTING FROM SPRAY APPLICATION TO A DORMANT PEACH ORCHARD

GLOTFELTY DWIGHT E SCHOMBURG CHARLOTTE J
MCCHESNEY MICHAEL M SAGEBIEL JOHN C SEIBER JAMES N

Interpretive Summary:
Occasionally illegal pesticide residues are found on crops to which the pesticide was never applied. How did the pesticide get onto the crop? The pesticide may have drifted from a neighboring field during spray application. Alternatively, the pesticide may have volatilized from the application site and been carried and deposited by rain, fog droplets or dust particles. As a first step in distinguishing between spray drift and other forms of pesticide loss from an application site an experiment was conducted to determine how much of a pesticide was lost from a dormant peach orchard during spray application and how much volatilized in the next few days after application. To accomplish this air samples were collected during spray application and for the next few days after application. Soil and tree rinse samples were also collected to determine how the spray was distributed in the orchard. Results showed while there is pesticide loss from the orchard during spray application, there is also continuing loss by volatilization after application. Also it seems more of the pesticide is found on the soil than on the trees in the orchard. From this preliminary work we have found may ways to improve these kinds of field measurements. This information will serve to help us in future experiments of this type and also other scientist doing these kinds of experiments.

Technical Abstract:
A preliminary experiment was conducted to determine the spray distribution, spray drift, and volatilization of diazinon applied in the conventional manner with an air-blast sprayer to a dormant peach orchard. Copper hydroxide and a dormant oil were applied along with the diazinon. Soil samples and tree rinse samples were used to determine the distribution in the orchard. Airborne losses were calculated by the integrated horizontal flux method from measurements of wind speed and pesticide concentration profiles obtained during and for several days following application. Diazinon was not distributed evenly between the trees and the soil in the orchard according to their relative areas. Most of the diazinon accounted for was found to be on the soil. The residue on the soil dissipated with a 19 day half life. Application drift losses were small compared to long-term volatilization losses, and we conclude that most of the diazinon in the Central Valley atmosphere during the dormant spray season results from volatilization. This result has important implications for designing strategies for controlling inadvertent contamination of other crops and the environment.

Submitted to: (approved 08/07/90)
CHEMOSPHERE

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THE EFFECT OF INOCULANT STRAIN AND ORGANIC MATTER CONTENT ON THE KINETICS OF 2,4-DICHLOROPHENOXYACID DEGRADATION IN SOIL

GREER LINDA E SHELTON DANIEL R

Interpretive Summary:
Soil bioremediation involves the use of microorganisms to detoxify and degrade pollutants in soil. In order for soil bioremediation to be successful, it is necessary that the appropriate strains be present or for the appropriate strains to be added to the soil (inoculation). In addition, it is critical that the microorganisms be able to survive and reproduce in the soil, and be able to degrade the pollutant at whatever concentration it may be present. In this study, the effect of choosing inoculant strains with different affinities for the herbicide 2,4-D on the rate of 2,4-D degradation in soil were examined. It was observed that at higher 2,4-D concentrations rates of degradation were comparable for two different strains, however, at lower concentrations the strain with the greater affinity for 2,4-D in liquid culture also degraded the 2,4-D faster and to a greater extent in soil. These results suggest that the choice of inoculant strain may be of considerable importance depending on the concentration of pollutant in soil. The effect of soil organic matter on rates of degradation were also investigated, since organic matter tends to adsorb pollutants, making them less available to microorganisms. As expected, rates of 2,4-D degradation in soils with high organic matter were slower than in soils with low organic matter using the same inoculant strain. These data demonstrate the importance of taking into consideration soil characteristics and pollutant concentrations when selecting an inoculant strain and in predicting rates of pollutant degradation.

Technical Abstract:
Rates of degradation of soluble and sorbed 2,4-D were monitored in low organic matter soil at field capacity amended with 1, 10, or 100 ug 2,4-D/g wet soil and inoculated with one of two bacterial strains (MI, 155) with similar maximum growth rates (Umax) but significantly different half saturation growth constants (Ks). Concentrations of soluble 2,4-D were determined by analyzing samples of pore water pressed from soil while sorbed 2,4-D was determined by solvent extraction. Sixty-five to seventy-five percent of total 2,4-D was present in the soluble phase at equilibrium, resulting in soil solution concentrations of ca. 8, 60, and 600 ug/ml 2,4-D, respectively. Soluble 2,4-D was metabolized preferentially, followed by degradation of both sorbed (after desorption) and soluble 2,4-D. Rates of degradation were comparable for both strains at soil concentrations of 10 and 100 ug/g 2,4-D, however, at 1 ug/g soil, 2,4-D was metabolized more rapidly by the strain with the lower Ks value (MI). Rates of biodegradation of soluble and sorbed 2,4-D were also monitored in high organic matter soil at field capacity amended with 100 ug 2,4-D/g wet soil and inoculated with the low Ks strain (MI). Ten percent of total 2,4-D was present in the soluble phase, resulting in a soil solution concentration of ca. 30 ug/ml 2,4-D. Rates of degradation in the high organic matter soil were slower than in the low organic matter soil, presumably, due to slower rates of desorption and microbial growth.

Submitted to: (approved 09/12/91)
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PHYSIOLOGICAL AND KINETIC COMPARISON OF SEVEN STRAINS OF 2,4-DICHLOROPHENOXYACETIC ACID DEGRADING BACTERIA

GREER LINDA E ROBINSON JOSEPH A SHELTON DANIEL R

Interpretive Summary:
Bioremediation is the process by which microorganisms are used to detoxify and/or degrade wastes and pollutants in the environment. In some instances the appropriate microorganisms may already be present at the site but in others it may be necessary to add or inoculate the site with the appropriate strains. In such situations it is necessary to select inoculant strains which will be able to survive and reproduce at the contaminated site, frequently under adverse environmental conditions. This requires that numerous pollutant degrading microorganisms be isolated and studied in order to develop a library of strains possessing a variety of different physiological and kinetic characteristics. Depending on the environmental characteristics of the site to be remediated, the most appropriate strain(s) can be selected for inoculation at that site. This study involves the isolation and characterization of several different strains of bacteria able to degrade the commonly used herbicide, 2,4-D, each with one or more unique characteristics which may be important in allowing that strain to survive and reproduce at a given site. This study demonstrates that diversity is common among naturally occurring microorganisms and that it should be possible to develop libraries of such pollutant degrading strains.

Technical Abstract:
Seven strains of 2,4-dichlorophenoxyacetic acid-degrading bacteria, including species of Pseudomonas, Alcaligenes and Bordetella, were compared on the basis of substrate range, mortality, and growth kinetics. Estimates of maximum growth rate (Umax, k1) and half saturation growth constant (Ks, k3) were obtained by fitting substrate depletion curves to a four parameter version of the integrated Monod equation. Estimates of Ks ranged from 2.2 ug/ml (10 uM) to 33.8 ug/ml (154 uM), while estimates of Umax ranged from 0.20 hr-1 (Td = 3.5 hr) to 0.32 hr-1 (Td = 2.2 hr). Estimates of Umax, but not Ks, were affected by changes in initial inoculum density. Maximum growth rates (Umax) were also estimated from turbidity measurements. Estimates of Umax ranged from 0.10 hr-1 (Td = 6.9 hr) to 1.0 hr-1 (Td = 0.7 hr). There was no correlation between estimates of Umax derived from substrate depletion curves and turbidity measurements (P=0.20). Mortality rates (27 C) varied dramatically between strains, with losses ranging from 90% in 28 days to greater than 4 orders of magnitude in 15 days. There was considerable variation in the ability of the seven strains to use different monomeric and polymeric carbon sources as growth substrates. Data from this study demonstrate there is significant physiological and kinetic variation in parameters likely to affect the survival and proliferation of 2,4-D degrading bacteria in the environment.

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MINERALIZATION OF PESTICIDE DEGRADATION PRODUCTS

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Interpretive Summary:
Alachlor (Lasso), metolachlor (Dual), paraquat (Gramoxone) and atrazine (Aatrex) solutions can be mineralized using a combination of chemical and biological treatments. Mineralization is the breakdown of a compound into its simplest elemental components, such as carbon dioxide, water, ammonia, nitrate, chloride and other salts. The pesticides were pretreated with either ultraviolet light or with ozone. The products of these chemical reactions were subsequently degraded by soil microorganisms. This work was conducted in an effort to reduce point source contamination to ground and surface waters stemming from improper disposal of pesticide wastes.

Technical Abstract:
Pesticides and pesticide degradation products, which are not mineralized to carbon dioxide, ammonia, water and inorganic salts, can leach and contaminate water supplies. Altering pesticides by photolysis or ozonation has been shown to enhance significantly the rate of mineralization. Photodegradation products of s-triazines, chloroacetanilides and paraquat were dechlorinated and/or oxidized. Ozonation of these same herbicides afforded products in which the alkyl side chains were oxidized or removed and the aromatic ring was oxidized or cleaved, however, dechlorination did not occur. In some cases, the matrix in which these compounds were found inhibited microbial activity.

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ACS SYMPOSIUM SERIES: FATE AND SIGNIFICANCE OF PESTICIDE DEGRADATION PROD.

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PHYSICAL AND CHEMICAL PROCESSES AFFECTING PREFERENTIAL FLOW

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Interpretive Summary:
Evidence has mounted over the past 10-20 years linking deeper than expected leaching of some agrichemicals into soils with preferred flow pathways. One major type includes macropores - large pores, fissures, channels, or other semi-continuous voids. These may be caused biologically, following decay of roots or through burrowing of earthworms or arthropods. Alternatively, macropores may form by natural soil aggregation. Fingering of soil water is a type of preferential flow caused by wetting front instability, and is most likely to occur in coarse-textured soils, especially at textural discontinuities. Nitrate, dye tracers, and pesticides of intermediate mobility have been shown to leach preferentially. For pesticides, transport through macropores is most likely when moderate to heavy rainfall occurs soon after application. Preferential flow appears to be more likely under long-term no-till conditions than with conventional till.

Technical Abstract:
Over the past 10-20 years evidence has mounted linking deeper than expected leaching of some agrichemicals into soils with preferred flow pathways. One major type includes macropores - large pores, fissures, channels, or other semi-continuous voids. These may be caused biologically, following decay of roots or through burrowing of earthworms or arthropods. Alternatively, macropores may form by natural soil aggregation. Fingering of soil water is a type of preferential flow caused by wetting front instability, and is most likely to occur in coarse-textured soils, especially at textural discontinuities. Nitrate, dye tracers, and pesticides of intermediate mobility have been shown to leach preferentially. For pesticides, transport through macropores is most likely when moderate to heavy rainfall occurs soon after application. Preferential flow appears to be more likely under long-term no-till conditions than with conventional till.

Submitted to: (approved 01/24/92)
PROC NAT SYMP PREFERENTIAL FLOW

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APPARATUS FOR STUDYING PESTICIDE DISSIPATION IN THE VADOSE ZONE

ISENSEE ALLAN R

Interpretive Summary:
Pesticides must leach through the vadose zone to reach groundwater. While much is known about pesticide degradation in the surface soil, comparatively little is known about their fate in the unsaturated zone. Subsurface-soil pesticide degradation studies are generally conducted under laboratory conditions, which may differ significantly from conditions in the vadose zone such as temperature, moisture, and oxygen. The apparatus described in this paper is designed to expose soils treated with pesticides to ambient vadose zone conditions for pesticide degradation studies. The apparatus consists of a casing and enclosed vadose exposure chamber (VEC). Soil cores placed in the VEC are in equilibrium with the surrounding soil. The system was tested for 100 days by measuring oxygen at 0.5, 1, and 2 m depths and comparing results to control samplers at 1 and 2 m depths. Nearly identical oxygen concentrations indicate that the apparatus provides a treatment chamber that is in equilibrium with the soil atmosphere and is isolated from the surface atmosphere. A major advantage of the apparatus is that radiolabeled pesticides can be used because the VEC is self-sealing in the event of unexpected flooding.

Technical Abstract:
An apparatus was constructed to study pesticide degradation in situ under vadose zone conditions. The apparatus consists of a 15-cm (i.d.) casing and an enclosed vadose exposure chamber (VEC) wherein pesticide treated soil cores, placed in the VEC, are in equilibrium with the surrounding vadose zone at selected depths. Vadose zone oxygen concentrations at 0.5, 1.0, and 2.0 m were monitored over 100 days and compared to oxygen concentrations in control samplers at 1 and 2 m depths. Nearly identical oxygen concentrations indicate that apparatus provides a treatment chamber that is in equilibrium with the soil atmosphere and is isolated from the surface atmosphere. Both radiolabeled and unlabeled pesticides can be studied in the apparatus because the VEC is self-sealing in the event of unexpected flooding

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LABORATORY APPARATUS FOR STUDYING PESTICIDE LEACHING IN INTACT SOIL CORES

ISENSEE A R
SADEGHI A M

Interpretive Summary:
Rainfall patterns (time between application and first rain event, intensity and duration), soil conditions, tillage practice and pesticide formulation are among the most important factors affecting pesticide leaching. Evaluation of these factors under field conditions is expensive, time consuming and subject to climatic variability. A laboratory method is needed to rapidly and accurately evaluate interactions between these factors as they impact pesticide leaching. This paper describes the design, construction, operation, and performance of an apparatus to study pesticide leaching through intact soil cores. A technique is described for mounting intact soil cores that nearly eliminates water movement at the soil-wall interface; a problem that often plagues soil column studies. A turntable is used to support and rotate up to 12 cores under a rain simulator capable of duplicating many rainfall rates. The capability for precise simulation of rainfall, accommodation of large numbers of soil cores and relatively fast set-up time make this apparatus ideal as a laboratory tool for conducting basic research.

Technical Abstract:
This paper describes the design, construction, operation, and performance of an apparatus to study pesticide leaching through intact soil cores obtained from no-till (NT) and conventional-till (CT) corn fields. A technique is described for rapidly mounting intact soil cores. A turntable is used to support and rotate soil cores (up to 12) under an oscillating rain simulator capable of producing rainfall rates of 1 to 30mm/h. Each soil core is attached to a filtration flask which is connected to a -10 to -20 Kpa vacuum supply. The CV of the rainfall delivery rate over a range of 2-12 mm/h averaged 3.7%. Dye studies using intact soil cores indicated slower water movement at the soil-wall interface than through the soil matrix. An experiment conducted to evaluate atrazine [2-chloro-4-(ethylamino)-6-(isopropylamino)-s-triazine] leaching through CT and NT soil cores indicated consistently greater leaching through CT cores and flawless operation of the apparatus. The capability of precise simulation of rainfall, accommodation of large number of soil cores, and ease of modification to meet a wide range of research parameters make this apparatus ideal for the laboratory evaluation of soil-water-pesticide interactions on pesticide leaching.

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JOURNAL OF ENVIRONMENTAL QUALITY

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THE GENES ENCODING S-TRIAZINE DEGRADATION ARE PLASMA ENCODED IN KLEBSIELLA PNEUMONIAE STRAIN 99

KARNS JEFFERY S
EATON RICHARD W

Interpretive Summary:
Plasmids are extrachromosomal, self-replicating pieces of DNA that often encode genes which impart unique properties upon the bacteria in which they reside. Because they can be transferred between bacteria plasmids are a means by which genetic information can be rapidly shared by members of the soil microbial community. Thus, plasmids can be a part of the mechanism by which soil microbes evolve the ability to degrade agricultural chemicals. This study shows that the genes which encode the enzymes for the degradation of the simple s-triazine compounds ammelide and cyanuric acid are carried on a large plasmid in a strain of Klebsiella pneumoniae. This plasmid is capable of transfer to other bacteria, indicating that it may act as an agent in the evolution of s-triazine degradation capabilities in bacteria. Ammelide and cyanuric acid are likely intermediates in the degradation of the herbicides atrazine, cyanazine and simazine.

Technical Abstract:
Klebsiella pneumoniae strain 99 degrades the s-triazine compound ammelide through cyanuric acid and biuret to yield urea, carbon dioxide and ammonia. The urea and ammonia formed from the degradation of ammelide or cyanuric acid are utilized as sources of nitrogen for growth of the organism. When plasmids of the IncI-alpha incompatibility group were transferred into K. pneumoniae strain 99 the ability to degrade s-triazine compounds was lost at high frequency. Analysis of the plasmid profiles of s-triazine positive and s-triazine negative derivatives of strain 99 indicated that the largest of the at least 5 plasmids present in this organism carried the genes encoding the s-triazine degradation pathway. Conjugal transfer of this plasmid into a type strain of Klebsiella planticola resulted in exconjugants able to utilize ammelide or cyanuric acid as nitrogen sources. Thus, all the genes required for s-triazine degradation are present on a large IncI-alpha plasmid in K. pneumoniae strain 99.

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SIMULATION OF ONE-DIMENSIONAL NITRATE TRANSPORT THROUGH SOIL AND CONCOMITANT NITRATE DIMINUTION

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Interpretive Summary:
The purpose of this study was to conduct prototype leaching experiments with one small, intact soil core to obtain information on the transport of nitrate in the presence and absence of a carbon source capable of supporting microbial growth. The size of the soil core selected was small enough to be readily extricated from the field, but large enough to preclude wall effects. Our soil core sample was taken from a riparian zone (RZ). Information obtained from the leaching experiments were used in a model by Parker and van Genuchten (1984) for solute transport and rates of solute decay and production during one-dimensional flow. This model accounted for experimentally determined transport and loss of nitrate. The model also provided transport parameter values for dispersion coefficient, nitrate exclusion factor, and a nitrate decay rate constant. Field managers who seek to optimize nitrate diminution in RZ's can use this approach to characterize nitrate transport characteristics in specific locations within an RZ before and after imposing management practices designed to increase the effectiveness of RZ's to lower the transport of nitrate. Other scientists can use this approach to obtain transport parameter values applicable to field dimensions by pooling information obtained from many such cores.

Technical Abstract:
A small core of intact soil provided information about the lateral transport and diminution of nitrate in the presence and absence of a carbon source capable of supporting microbial growth. The core (6.2 cm in diameter by 15 cm in length) was obtained by pushing a plastic cylinder horizontally into the wall of a trench excavated in a riparian zone where water moves laterally. Under conditions favorable for nitrate diminution, pulses of nitrate solution containing carbon were passed through the core and leached with water. Effluents were collected at fixed intervals and analyzed. Nitrate losses ranged from 4% to 72%, presumably via denitrification. Effluent nitrate concentrations and pore water velocities were entered into a model developed by Parker and van Genuchten for one-dimensional convective-dispersive solute transport and for solute decay and production. The model accounted for the experimentally determined transport and loss of nitrate (R^2 = 0.99). The model also provided values for dispersion coefficient, nitrate exclusion factor, and nitrate decay rate constant. Nitrate loss was reasonably accounted for by a decay rate constant (avg. = 2.21/day) that increased as observed nitrate loss increased. A sizable factor for anion exclusion and/or the presence of immobile-water regions was necessary to account for the rapid passage of nitrate through the soil core. Peclet numbers, which are inversely related to dispersion coefficient, were low. These low numbers and a retardation factor of less that 1 suggested that the flow through the soil core may have been heterogeneous.

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UGA IS NATURALLY SUPPRESSED IN WILD-TYPE BACILLUS SUBTILIS

LOVETT P S AMBULOS N P JR
MULBRY WALTER W NOGUCHI N ROGERS E J

Interpretive Summary:
The biochemical mechanisms that underlie the genetic processes of bacteria and more complex organisms are the focus of intense study worldwide. Within this realm of research are studies of the processes by which genetic information encoded on the DNA of an organism is deciphered so that such cellular material as proteins are made. Recombinant DNA techniques were used to demonstrate that the 'genetic punctuation' used to mark the location on DNA that protein synthesis should stop varies between two different important groups of bacteria. This study revealed that a DNA sequence, which functions as a genetic punctuation signal in many such organisms as the well studied intestinal bacteria Escherichia coli, is naturally ignored in the industrially important bacteria belonging to the group Bacillus. This finding is important to scientists who examine DNA while searching for important genes and to those who are trying to manipulate important genes so that they may be expressed in useful organisms.

Technical Abstract:
The ochre codon UAA functions as a translation termination codon in Bacillus subtilis. Mutations in B. subtilis which suppress the ochre codon were found to insert lysine (sup-3 and sup-67) or leucine (sup-44). As with the ochre codon, the opal codon UGA, occurs at the end of several coding sequences that function in B. subtilis but not within coding sequences. This location is consistent with the assignment of UGA as a stop codon. However, if UGA were only a stop codon it should be expected that the extensive generation of random mutations in B. subtilis during the last three decades would have resulted in the insertion of UGA within a coding sequence. This class of mutation would have been revealed by the subsequent isolation of an extragenic suppressor, which predictably would fail to suppress UAA/UAG. Since all known nonsense suppressor mutations of B. subtilis suppress ochre and amber mutations these cannot suppress UGA. Thus either UGA has never been introduced into a coding sequence in B. subtilis, which is highly unlikely, or B. subtilis naturally suppresses UGA. By use of mutations within the cat-86 gene we show that UGA is naturally suppressed in each of four strains of B. subtilis. Weak suppression is also observed in Staphylococcus aureus. We demonstrate that in B. subtilis the suppression of UGA is due to insertion of tryptophan. The apparent efficiency of UGA suppression in B. subtilis ranges from about 1 to 6%.

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JOURNAL OF BACTERIOLOGY

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EPTC DEGRADATION BY ISOLATED SOIL MICROORGANISMS

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DICK W A
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Interpretive Summary:
Pre-emergence carbamothioate herbicides such as EPTC (EPTAM or Eradicaine) are important tools for weed control in corn. The efficacy of these herbicides has been shown to be adversely affected by rapid degradation of the herbicide such that sufficient residues do not remain to control weeds. This rapid degradation phenomenon has become known as 'enhanced' or 'accelerated' pesticide degradation and soils in which this occurs are known as 'agressive' or 'problem' soils. In many cases the rapid degradation of a pesticide in a problem soil has been shown to be due to the presence of microbes that are capable of degrading the pesticide at a rapid rate. We have isolated several bacteria that are able to rapidly degrade EPTC. The EPTC degradation capability was rapidly lost by these bacteria, suggesting that plasmid DNA might encode the EPTC degradative enzymes in these cell but we were unable to implicate any of the three plasmids in these strains in the degradation of EPTC. Studies with EPTC labeled with radioactive carbon in different parts of the molecule suggested that the initial attack on the EPTC molecule was hydrolysis of the carbamate ester followed by degradation of the hydrolysis products. This information will aid further studies into the biochemical mechanisms of EPTC degradation by bacteria and into the genetic mechanisms by which a soil microbial community adapts to accomplish the rapid degradation of herbicides.

Technical Abstract:
Microorganisms capable of degrading the herbicide EPTC (s-ethyl-N,N-dipropylthiocarbamate) were isolated from three soils with and without histories of carbamothioate use. All EPTC-degrading isolates belonged to the genus Rhodococcus. All three isolates in pure culture systems degraded 50 ug mL-1 of technical EPTC in as little as fourteen hours at low cell densities, and used the molecule as a sole source of carbon and energy. Growth of the isolates in rich media led to the frequent loss of EPTC-degrading ability although plasmids encoding for EPTC degradation could not be identified. 14-C-labeled EPTC experiments suggested that the degradation of EPTC proceeds by initial attack at the carbonyl linkage, followed by degradation of the dipropylamine sidechain.

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JOURNAL OF AGRICULTURAL FOOD CHEMISTRY
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EVALUATION OF THE PRE-SIDEDRESS NITRATE SOIL TEST IN MARYLAND

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Interpretive Summary:
Society is concerned about nitrate enrichment of groundwater and estuaries. The agriculture community shares this concern and is actively researching ways to improve nitrogen use efficiency. A basic tool to increase nitrogen efficiency is a nitrogen soil test. A nitrogen soil test for corn was evaluated in Maryland which measures the nitrate nitrogen concentration in the top foot of soil when the corn is 6-12 inches tall i.e., about two weeks before planned sidedressing. The test was evaluated on research farm experiments over a wide range of soils and over several growing seasons. These tests studied the effects of various tillage systems, applications of manure or composted sludge, winter cover crops, and applications of fertilizer. The results show that the soil test can identify nitrogen sufficient sites, i.e. sites needing little or no sidedress fertilizer nitrogen. Nitrogen sufficient sites contained greater than 22 parts per million nitrate nitrogen and were usually associated with previous inputs of manure or composted sludge, or had grown a legume cover crop. The test will help nutrient consultants, extension agents, and farmers identify nitrogen sufficient sites and thereby: conserve fertilizer nitrogen, improve nitrogen use efficiency, and reduce nitrate losses to the environment.

Technical Abstract:
Nitrate enrichment of groundwater and estuaries is a major concern in the Mid-Atlantic States. The pre-sidedress nitrate test (PSNT) was evaluated for corn in Maryland by collecting 0-30 cm soil samples when the corn (Zea mays L.)was 15-30 cm tall, determining NO3-N and (NO3+NH4)-N by steam distillation, and measuring corn grain yield. The replicated research farm experiments were located in both the Piedmont and Atlantic Coastal Plain regions and included 7 soil types and 5 growing seasons. A total of 47 treatment-year combinations were studied including variables of tillage, organic amendments, cover crops, and fertilizer N rates. The PSNT accurately reflected differences in N availability to corn due to prior manure applications (poultry manure or dairy manure), prior composted sludge applications, and prior N inputs from incorporated winter cover crops. The PSNT can be used on either no-tillage or plow-tillage systems. Soil NO3-N concentrations greater than 22 mg/kg soil, or (NO3+NH4)-N concentrations greater than 27 mg/kg soil, were associated with relative yields of 95% or higher. At soil N concentrations less than the above values the variation in relative yield was considered to be too large to use the test for quantitative prediction. The PSNT successfully identified N sufficient sites across a range of soil textures and drainage classes. By identifying N sufficient sites, the PSNT will help farmers conserve fertilizer N and reduce NO3-N losses to the environment.

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Technology Transfer Automated Retrieval System - (TEKTRAN)

EFFECT OF COVER CROPS ON GROUNDWATER QUALITY

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Interpretive Summary:
Farmers face a difficult task as they seek to balance the competing goals of maintaining farm profitability, by ensuring an adequate supply of N to crops, yet avoiding excessive N rates that could increase nitrate losses to groundwater. Winter cover cropping is an important management practice that can reduce nitrate leaching into groundwater by: using water to reduce percolation, assimilating nitrogen to reduce soil nitrate, and through synchronized competition between the cover crop N uptake season vs the nitrate leaching season. A summary of the literature clearly shows that cover cropping can reduce nitrate leaching by 20 to 80% compared to no cover cropping. Furthermore, grass cover crops are two to three times more effective than legumes (hairy vetch, clovers, etc.) in reducing nitrate leaching. The value of cover cropping will be greatest in the Southeastern U.S. and in irrigated agriculture, but model simulation results predict that cover cropping will have a broadly beneficial impact over much of the U.S. Management practices which will improve nitrate conservation by cover cropping include selection of a species with vigorous fall growth and early planting. This review and analysis will be important to applied research scientists, extension agents, and soil conservationists as they plan and advise farmers on development of management systems to reduce nitrate losses into groundwater.

Technical Abstract:
Winter cover crops can influence nitrate leaching and groundwater quality by: i) influencing the water budget ii) affecting the soil nitrate concentration, and iii) through synchronized competition during the water recharge season. Experimental results from the literature clearly show that cover crops can reduce both the mass of N leached, and the nitrate concentration of the leachate, by 20 to 80% compared to no cover crop. The grasses and brassicas are two to three times more efficient than legumes in reducing N leaching. Management factors which improve N conservation are selection of a species with vigorous fall growth and early cover crop establishment. Using the EPIC model it was estimated that a winter cover crop will have the greatest impact on nitrate leaching in the humid Southeast and in irrigated agriculture, but cover crops had a positive effect for all scenarios evaluated. Additional research needs include: more direct field measurements of nitrate leaching for a range of soils, cover crops, and climates; improved understanding and modeling of the N cycle; and improved plant germplasm for use as cover crops.

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SOIL & WATER CONSERVATION SOC INTERNAT CONF ON COVER CROPS FOR CLEAN WATER

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USING WINTER COVER CROPS TO RECYCLE NITROGEN AND REDUCE LEACHING

MEISINGER JOHN J
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Interpretive Summary:
Farmers face a difficult task as they seek to balance the competing goals of maintaining farm profitability, by ensuring an adequate supply of N to the crop, yet avoiding excessive N rates that could degrade groundwater quality. Both legumes and grasses have been utilized in the Southeast as winter cover crops but their ability to recycle corn fertilizer N has not been evaluated. We directly measured the ability of grass (cereal rye and ryegrass) vs, legumes (hairy vetch and crimson clover) to recycle corn fertilizer N by adding isotopically tagged fertilizer to corn and measuring the uptake of the corn fertilizer by the covers. Our field results show that grasses are clearly superior to legumes in retaining corn fertilizer N with grasses taking up 50-60% of the excess fertilizer (in tops plus roots) while legumes took up less than 10%. These field results agree very well with earlier lysimeter results which also showed that grasses were superior to legumes at conserving N. The use of grass winter cover crops is an old practice which should be re-incorporated into modern cropping systems.

Technical Abstract:
Winter cover crops are a management practice which can reduce nitrate leaching by i) utilizing water for growth and thereby reduce percolation, and ii) absorbing N to meet nutritional needs. Both legumes and grasses are utilized as cover crops in the Southeast but their ability to recycle fertilizer N in the field has not been evaluated. We directly measured the ability of grass vs. legume cover crops to retain corn fertilizer N by adding 15N labelled fertilizer to corn, subsequently growing grass and legume covers, and measuring the uptake of the corn fertilizer N by the covers. We found that grasses (cereal rye and ryegrass) retained 50-60% of the corn fertilizer N in their tops plus roots, while legumes (hairy vetch and crimson clover) retained 10% or less. These recent field results agree with earlier lysimeter results in the Southeast which shows that nitrate leaching with grasses was reduced 75-85% compared to the no-cover controls, while the corresponding value for legumes was 0-12%. Agriculture scientists should therefore place greater emphasis on incorporating grass cover crops into modern cropping systems.

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PURIFICATION AND CHARACTERIZATION OF THE METHYLCARBAMATE HYDROLASE FROM PSEUDOMONAS STRAIN CRL-OK

MULBRY WALTER W
EATON RICHARD W

Interpretive Summary:
Many studies have demonstrated that a variety of pesticides are degraded by enzymes produced by soil bacteria. One area of great interest in biotechnology is the use of these bacterial enzymes in degrading pesticide wastes. Some pesticides are effectively detoxified by a single enzymatic reaction, while others require a series of reactions before they are rendered nontoxic. A bacterial enzyme that hydrolyzes and detoxifies the methylcarbamate pesticide carbaryl was isolated and characterized in this study. While this enzyme was similar to a previously identified carbamate hydrolase, it differed from the known enzyme with respect to its size and substrate specificity. Further comparison of these enzymes may lead to a precise understanding of how they function and will aid efforts aimed at modifying such enzymes to increase their activities and substrate ranges.

Technical Abstract:
Pseudomonas sp. strain CRL-OK was isolated from sewage sludge by enrichment using the insecticide carbaryl (1-naphthyl N-methylcarbamate) as a carbon source. A unique cytosolic enzyme that hydrolyzes the carbamate linkage of carbaryl was purified more than 1000-fold from cell-free extracts of strain CRL-OK. After purification, activity was stable for greater than one month at 4 C and stable indefinitely at -80 C. The hydrolase is composed of two identical subunits of 85,000 daltons and has temperature and pH optima of 60 C and 8.5, respectively. Its substrates include the N-methylcarbamate pesticides carbofuran and aldicarb, but not the phenylcarbamate CIPC, the thiocarbamate EPTC, nor the dimethylcarbamate o-NPDC. Hydrolase activity was not affected by the ionic detergent SDS at concentrations up to 0.2%, by 2-mercaptoethanol concentrations up to 0.2mM, nor by prolonged incubation in the presence of the divalent cation chelator EDTA.

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JOURNAL OF APPLIED AND ENVIRONMENTAL MICROBIOLOGY

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SPATIAL AND TEMPORAL VARIABILITY OF CARBOFURAN DEGRADATION IN SOIL

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SHELTON DAN R

Interpretive Summary:
Microbial degradation is an important fate of pesticides in soil. Thus, prediction of the fate of pesticides in the environment requires knowledge of the factors which control microbial degradation rates. This necessarily requires determination of the spatial and temporal variability associated with pesticide degradation rates in soil. Our study was designed to quantify the spatial and temporal variability associated with microbial degradation of the insecticide, carbofuran. We observed higher rates of carbofuran degradation were associated with samples collected in the row as compared to samples collected between corn rows. Soil water content and microbial biomass jointly influenced the kinetic patterns observed as well as the spatial variations of carbofuran degradation activity.

Technical Abstract:
Loss of pesticide efficacy resulting from enhanced rates of microbial degradation has been observed with several pesticides including the insecticide carbofuran. Soils in which this phenomenon occurs are often referred to as "problem soils". There have been few studies of the spatial or temporal variability of carbofuran degradation. Our study was designed to evaluate the spatial variability of carbofuran degradation activity in a conventional-till and a no-till corn field, and to assess temporal variations of carbofuran degradation activity. Soil samples were collected at two positional locations in each field (in-row and between-row) at three times during the growing season. Within the planting furrow maximum rates of carbofuran degradation were higher and resulting half-lives of carbofuran (DT-50%) were lower than in samples collected between corn rows. Interactive effects of both microbial biomass and soil water content appeared to influence spatial variations in the degradation kinetics of carbofuran as well as the positional differences observed. Temporal variations in carbofuran degradation appeared to be dominated by soil water content. At this time if remains uncertain whether the observed increase in numbers of carbofuran degrading organisms, in the row, was in response to the banded application of carbofuran, or to increased carbon availability in the rhizosphere.

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EVALUATION OF METHODS FOR CHARACTERIZING CARBOFURAN HYDROLYSIS IN SOIL

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Interpretive Summary:
Recent concerns over groundwater and surface water pollution has demanded that current agricultural practices be modified. Effective mitigation of the environmental consequences of pesticide application to agricultural lands, either through modification of agricultural management practices or pesticide formulations, will require an increased quantification of the fate of pesticides applied to soils. Degradation by microorganisms has been documented as a predominant fate of pesticides in soil; however, efforts to precisely quantify and/or predict pesticide degradation rates is hampered by high spatial and temporal variability associated with microbial processes in nature. The objective of this work was to design and evaluate a protocol for assessing pesticide degradation rates in soil, using the insecticide, carborfuran, as a model compound. Two factors were considered in the methods development: i) the mode of soil handling and pesticide application, and ii) the method of data summarization.

Technical Abstract:
The objective of this study was to develop a method to investigate the soil/environmental factors influencing the spatial and temporal variability of carbofuran hydrolysis in field soils. Three soil treatment modes were evaluated: i) sieved soil/sprayed pesticide application, ii) injected pesticide application/sieved soil, and iii) injected pesticide application/intact core incubation. This last method was developed to more closely mimic field condition in which high localized concentrations of carbofuran occur due to the banding of granular material at planting time. Several mathematical models for describing sigmoidal product appearance data were evaluated and a general saturation model was found to yield the best fit. Using parameter estimates obtained from this model in statistical tests were found that the intact core soil treatment yielded significantly longer half lives for carbofuran degradation. Additional experimental evidence suggests that the soil structure of the intact cores retarded diffusion of the 14CO2 produced from carbofuran degradation which influenced the kinetic pattern observed.

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SOIL SCIENCE SOCIETY OF AMERICA JOURNAL

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CONTAMINATION OF GROUNDWATER BY ATRAZINE AND SELECTED METABOLITES

H. B. PIONKE
D. W. GLOTFELTY

Interpretive Summary:
The widespread use of atrazine and its conversion to a metabolite (DEA) that is more persistent, more leachable and as phytotoxic has raised concerns regarding groundwater contamination. We show widespread contamination, but at very low concentrations which suggest little environmental impact. However, the contamination patterns in time and space, and their stability in the more contaminated well waters, suggest that degradation rates, leaching rates and the atrazine/DEA storage together control the DEA concentrations in groundwater. Dilution, usually dominant in groundwater systems, has little affect. Thus, the atrazine-metabolite relationships developed here for soil-groundwater indicate this to be a useful model for examining the behavior and patterns of other pesticides/metabolite combinations as they move from soil to groundwater to stream baseflow.

Technical Abstract:
Groundwaters from an agricultural PA watershed were analyzed for atrazine, cyanazine, simazine, and the desethylated (DEA), and desisopropylated (DIA) atrazine metabolites. Atrazine and both metabolites were found in most groundwaters including deep wells, a spring and groundwaters about to become streamflow. The highest concentrations of atrazine and the dominant metabolite, DEA, were found in groundwaters draining areas dominated by corn production, especially after the first major groundwater recharge period following herbicide application.

Submitted to: (approved 06/22/90)
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Technology Transfer Automated Retrieval System - (TEKTRAN)

SPINACH LEAF CHLOROPLAST NO2- PHOTOREDUCTION IS STIMULATED BY FRUCTOSE-1,6-BISPHOSPHATE AND/OR TRIOSE PHOSPHATES

ROBINSON J MICHAEL

Interpretive Summary:
It well known that crop plant leaf nitrogen assimilation requires the formation of carbon metabolites in order to use these compounds for the insertion of ammonia to form amino acids. There is some evidence which indicates that enzymes associated with leaf carbon metabolism are stimulated by the onset of primary nitrogen metabolism. However, what has not been clear is whether or not that products of carbon metabolism, triose and hexose phosphate intermediates, stimulate enzymes associated with the conversion of nitrate to ammonia. However, a Beltsville Area Plant Physiologist has observed and confirmed that compounds generated during the primary steps of photosynthetic CO2 assimilation are stimulatory with respect to the chloroplast enzyme nitrite reductase. This observation is important to crop plant leaf photosynthetic nitrogen assimilation, because it indicates that there are photosynthetically generated carbon compounds which deliver a metabolic signal to the chloroplast N assimilatory enzymes indicating that there are additional carbon skeletons being produced in the light. These compounds ultimately serve as amino group acceptors. Genetic engineers may be able to take advantage of these observations in order to better understand how to improve the efficiency of enzymes associated with foliar primary nitrogen assimilation.

Technical Abstract:
Nitrite photoreduction (NiPR) in chloroplasts is catalyzed by the rate-limiting, reduced ferredoxin driven enzyme, nitrite reductase (NiR). Since chloroplastic NiR is coupled in linear sequence with chloroplastic glutamine synthetase and glutamate synthase (also driven by reduced ferredoxin), then the ultimate result of NiPR activity is the light dependent formation of additional glutamate for export from the chloroplast into the cytoplasm. In intact spinach leaf chloroplast isolates, NO2- photoreduction (NiPR) often is enhanced (18-30%) in the presence of CO2 fixation (CPA). It was found that this stimulation of NiPR (nitrite reductase), observed during CPA, appeared to be mediated by fructose-1,6-bisphosphate (FBP) glyceraldehyde-3-phosphate (GAP) and/or dihydroxyacetone phosphate (DHAP) apparently generated during CPA. GAP and DHAP are precursors of 3 carbon metabolites, e.g. pyruvate (Pyr), in the foliar anaplerotic pathways. These metabolites serve as precursor carbon skeletons for amino acids. For example, Pyr can be converted to alanine mediated through transamination from glutamate. Thus, the stimulation of NiR by FBP, GAP and/or DHAP may be a reflection of a metabolic signal indicating to the chloroplast N assimilatory sequence that there are additional carbon skeletons being produced in the light which ultimately serve as amino group acceptors.

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CUR TOP IN PLANT BIOCHEM & PHYSIOL 1990

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EFFECT OF TILLAGE SYSTEMS AND RAINFALL PATTERNS ON ATRAZINE DISTRIBUTION IN SOIL

SADEGHI A M
ISENSEE A R

Interpretive Summary:
Atrazine is a widely used herbicide for the control of weeds in corn production. Variable levels of atrazine residues in soil of the rooting zone of corn and shallow groundwater have been reported under various agricultural tillage systems. The two-year (1987 & 1988) field data were analyzed to evaluate the effects of conventional and no-till practices, and their interactions under different climatic regimes on the distribution and leaching characteristics of atrazine in corn production. Overall, atrazine residues within the top 10 cm soil depth of conventional-till plots were higher than in the no-till plots, regardless of the difference in the rainfall patterns. The approximately two fold higher mean atrazine residues in the conventional-till plots over no-till plots in 1988 were most likely related to the rainfall that began 12 h after application. In contrast, in 1987, it did not rain until 3 to 4 days after application and the differences were not so great.

Technical Abstract:
High variability of atrazine residues in soil and shallow groundwater have been reported under various agricultural management systems. This two-year study was conducted to evaluate atrazine residue levels in soil as influenced by no-till (NT) vs conventional-till (CT) under natural rainfall conditions. Atrazine was applied annually at the rate of 1.34 kg/ha to two NT and two CT plots one day after planting corn. Atrazine residues within the 0-10 cm soil depth of CT plots were higher than in the NT plots, regardless of the difference in the rainfall patterns. Higher (ca. 61%) mean atrazine residues in the CT plots than NT plots in 1988 was most likely related to the rainfall that began 12 h after application. In contrast, in 1987, it did not rain until 3 to 4 days after application and the residues in the CT were only 31% higher than in NT. These results indicate that even a subtle difference in the temporal rainfall distribution can result in a marked effct on the spatial distribution of atrazine.

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JOURNAL OF ENVIRONMENTAL QUALITY

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EVALUATION OF A TWO DIMENSIONAL HORIZONTAL FLOW SYSTEM

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Interpretive Summary:
Field studies have shown that concentrations of agricultural chemicals are reduced significantly as the chemicals move laterally from the edge of the field through riparian zones to nearby streams and estuaries. A laboratory-scale chamber was designed and tested as a prototype for constructing a larger field-scale chamber to characterize in-situ levels and transformations of agrichemicals within the riperiam zone areas. The multi-port injection technique that we have developed and tested in these studies promises to provide a useful experimental technique as a preliminary to studying horizontal flow processes in field.

Technical Abstract:
Results of several field studies have shown that a significant reduction in concentration of agricultural chemicals(agrichemicals) occurs as they move laterally from the edge of the field through riparian zones (RZ) to nearby streams, estuaries, etc. A laboratory-scale chamber was designed and tested as a prototype for constructing a larger field-scale chamber to characterize in-situ levels and transformations of agrichemicals within the RZ areas. The chamber was made of plexiglas with dimensions of 1.2 by 0.6 by 0.3 m and filled with fine sand. A multi-port arrangement (6 rows and 12 columns of 1-cm diameter holes) on both end walls of the chamber provided a plate-like combination for uniform solute application at the inlet side and allowed sampling from all ports at one time at the outlet side. An equilibrium-base convection-dispersion model was applied to the Cl breakthrough data of each of the 50 outlet ports in order to visualize the 2-dimensional distribution of pore water velocity (V) and dispersion coefficient (D) values at the outlet plate. The spatial variability of V and D were attributed to the heterogeneity in the packing process, evaporation losses from the sand surface, and density difference of the miscible displacement solutions.

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SOIL SCIENCE SOCIETY OF AMERICA JOURNAL

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ROOT EXTRACTION OF NUTRIENTS ASSOCIATED WITH LONG-TERM SOIL MANAGEMENT

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Interpretive Summary:
One of the main challenges facing agricultural research at the present time, is to identify farming practices that will maintain long-term soil fertility and crop yields, while at the same time allowing a reduction in the amount of fertilizer chemicals added and number of tillage operations. From our current knowledge of processes determining the uptake of nutrients by crops, several areas of research are needed to meet this challenge. These areas of research involve determining the effect of tillage practice, fertilizer type and placement, residue management, and crop selection on the amount of water and plant available nutrients in the volume of soil where roots are actively growing. Results from this research should enhance the sustainability of agricultural production systems.

Technical Abstract:
One of the main challenges of agricultural research is to identify management practices that maintain long-term soil fertility and crop production with reduced chemical inputs and tillage operations. This paper reviews the current state of knowledge and research needs concerning the impact of soil management on the root extraction of nutrients. Manageable variables controlling soil-water content, nutrient availability, root growth and development, and thereby, root extraction of nutrients are interactive, complex, and dynamic. There is, thus, a need for team research involving physical, chemical, and biological disciplines. This should focus on tillage practice, fertilizer type and placement, residue management, and crop selection to coincide the positional availability of soil water and nutrients during periods of active root growth and nutrient uptake. It is also important to determine the relative importance of soil properties on organic matter cycling as influenced by soil fauna and flora, tillage, and crop rotation. Information from identified research needs and integration of existing knowledge into management systems should facilitate progress towards enhancing sustainable soil fertility.

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ADVANCES IN SOIL SCIENCE

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EFFECT OF MOISTURE AND BIODEGRADATION OF CARBOFURAN IN SOIL

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Interpretive Summary:
Pesticides are generally perceived to pose a threat to the environment because of their persistence. This is due to the fact that most pesticides are not readily degraded by soil microorganisms. Some pesticides, however, are so rapidly degraded by microorganisms that they fail to control the target pest, resulting in a loss of efficacy. Carbofuran, an insecticide used to control the corn root worm is one such pesticide. This study was undertaken in an effort to better understand those environmental facts which control the rate at which carbofuran is degraded. Soil moisture is an important controlling factor because it effects both the activity of the soil microorganisms and the availability of carbofuran for degradation. The result of this study indicates that decreasing soil moisture severely depresses rates of carbofuran degradation. This is primarily due to the inhibitory effect of desiccation on the carbofuran degrading microorganisms, as opposed to carbofuran availability. The factors which control rates of pesticide degradation in soil, however, are very complex.

Technical Abstract:
Rates of degradation of soluble and sorbed 14C-carbonyl-carbofuran (4 ug/g soil) as well as evolution of 14CO2 were monitored in soil incubated at initial moistures of 20% (-0.4 bar), 17.5% (-0.8 bar), 15% (-1.6 bar), 12.5% (-3.4 bar), 10% (-7.0 bar) and 7.5% (-15 bar). Concentrations of soluble carbofuran were determined by analyzing pore water pressed from soil while sorbed carbofuran was determined by solvent extraction. Rates of degradation were comparable at -0.4 and -0.8 bar with degradation of greater than 97% of the carbofuran within 2 wk, but rates decreased with diminishing moisture content and only 7% was recovered 14CO2 after 5 wk incubation at 7.5% moisture (-15 bar). All curves were sigmoidal, which indicated of microbial growth. Concentrations of soluble carbofuran initially decreased and sorbed carbofuran increased as adsorption occurred. This was followed by losses of both soluble and sorbed carbofuran as degradation proceeded. Kd values (ug carbofuran per g soil/ug carbofuran per ml soil solution) increased during incubation but at progressively slower rates as a function of decreasing soil moisture. Kd values in previously air-dried (abiotic) soils incubated at -0.6 bar were 0.12-0.13 after 2 days incubation but increased to 0.16-0.17 after 3 wk. Additions of carbofuran 2 wk prior to or at the time of wetting of the air-dried soil had no effect on rates of adsorption. These data indicate that soil moisture can effect rates of biodegradation either by inhibition of microbial activity (desiccation) or by effecting substrate availability (diffusion and adsorption). This study is consistent with previous studies which indicate that: 1) only compounds in soil solution are available for metabolism and 2) adsorption/desorption is a two site/two step process.

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JOURNAL OF AGRICULTURAL AND FOOD CHEMISTRY

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COMPARISON OF VARIOUS WINTER COVER CROPS' ABILITY TO RECYCLE CORN FERTILIZER NITROGEN IN MARYLAND

SHIPLEY PAUL R MEISINGER JOHN J DECKER A MORRIS

Interpretive Summary:
Grasses are about four times as efficient as legumes at recycling excess nitrogen fertilizer. Excess nitrate from fertilizers can leach down to groundwater and become a pollutant. One way farmers try to avoid this problem is by planting a crop in late fall that will use up any nitrate not used by the last crop. The question then becomes: Which crops are best to use as winter cover crops? For a clear--if regional--answer, Agricultural Research Service scientists used an ammonium nitrate fertilizer that was tagged with N-15, an isotopic form of nitrogen that is found only in minute quantities in nature or in commercial fertilizer. At a University of Maryland research farm on Maryland's Eastern Shore, ARS soil scientist J. J. Meisinger and graduate student Paul Shipley applied the tagged fertilizer to corn for 2 years. Each fall they measured the residual tagged nitrogen in the soil to 2 feet and planted cover crops of cereal rye, ryegrass, hairy vetch, and crimson clover. The next spring the cover crops were harvested, ground, and analyzed for N-15. Any N-15 found could come only from the previous corn fertilizer. Considering only the aboveground nitrogen uptake, they found cereal rye and ryegrass were the best winter cover crops by far. Each took in about 40 percent of the fall residual nitrate. The hairy vetch and crimson clover legumes each took up only about 10 percent.

Technical Abstract:
Autumn residual fertilizer nitrogen (FN) is vulnerable to leaching loss in humid areas and represents a wasted resource and a potential pollutant. Legume and grass winter cover crops were evaluated for their ability to assimilate residual FN as a means of reducing leaching after corn. Labelled FN (15N depleted) was applied to corn in 1986 and 1987 at rates of 0, 168 and 336 kg N/ha. After corn harvest cover crops of, hairy vetch (Vicia villosa Roth), crimson clover (T. incarnatum L.), cereal rye (Secale cereal L.), annual ryegrass (Lolium multiflorum) or native weeds were established. Harvests were made three times the following spring; dry matter yields, %N, and atom % 15N were determined to assess FN uptake. Average FN uptake over the 2 years by the cereal rye, annual ryegrass, hairy vetch, crimson clover and native weed cover on the 168 kg N/ha treatment was 11, 6, 3, 4 and 2 kg N/ha respectively, at the second harvest (mid April). Corresponding FN uptake on plots previously fertilized with 336 kg N/ha were 48, 29, 9, 8 and 6 kg N/ha (L.S.D. 0.05=7 kg FN/ha). Fall FN levels in the soil averaged 17 and 114 kg N/ha over both years for the 168 and 336 kg N/ha rates, respectively. These results indicate that with high residual N the grass cover crops conserved the most FN, with cereal rye being the most efficient recycler through mid-April.

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AGRONOMY JOURNAL

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ENZYME-BASED STRATEGY FOR TOXIC WASTE TREATMENT AND WASTE MINIMIZATION

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Interpretive Summary:
Biotechnology promises to provide new and innovative processes for the disposal of waste agrochemicals. In some cases, the use of enzymes in waste treatment processes may have advantages over the use of whole organisms. Enzymes derived from recombinant organisms should have fewer regulatory hurdles to clear before they can be used. This is, in part, due to the fact that enzymes do not replicate and hence have finite lifetimes once applied. Before an enzyme can be used in any process the factors affecting the rate and extent of its degradation of the target compound must be determined. This study used the degradation of the insecticide coumaphos in waste cattle dips by parathion hydrolase as a system for the modeling of an enzyme-based waste treatment scenario. Parathion hydrolase was shown to be able to selectively remove a toxic metabolite of coumaphos, potasan, when enzyme concentrations were limited. This was shown to be due to potasan's increased solubility in water. Mathematical models considering enzyme half-life, temperature, and coumaphos and potasan concentrations, were derived which allown end users to determine how much enzyme needs to be added to any volume of cattle-dip to accomplish either the elimination of potasan from material still being used or the elimination of both potasan and coumaphos from waste material.

Technical Abstract:
The increasing amounts of pesticides used throughout the world as well as the increasingly stringent governmental regulations concerning waste disposal mandates improved techniques of waste disposal and minimization. In this paper, parathion hydrolase, an enzyme with proven effectiveness at hydrolyzing organophosphates, was used to treat a cattle-dipping liquid containing the pesticide coumaphos, which is used to kill a disease-causing tick. Waste is generated from this process when a toxic dechlorination product of coumaphos, potasan, accumulates to concentrations hazardous to the cattle. This pesticide system was used as a model to demonstrate how enzyme technology can be applied to waste treatment and minimization. Kinetic experiments showed that the hydrolysis of the two organophosphate substrates can be modeled as first order reactions with identical rate constants. It was further shown that the enzyme is capable of hydrolyzing only dissolved substrates. Because of the eight-fold greater solubility of potasan than coumaphos (16.9 vs 2.2 umol/L) it was possible to utilize the enzyme to selectively hydrolyze potasan. Thus, by limiting the amount of enzyme it is possible to selectively remove potasan to extend the lifetime of the cattle-dipping liquid, thereby reducing the amount of waste generated. Based upon experimental results, a mathematical model describing the system was developed and verified. The mathematical model was then used to simulate the ability of the enzyme to hydrolyze the total amount of organophosphates, and to selectively degrade all of the toxic potasan without a significant loss of coumaphos.

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BIOTECHNOLOGY AND BIOENGINEERING

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INFLUENCE OF SAMPLE VOLUME ON NITRATE-N MEASUREMENTS

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Interpretive Summary:
The volume of soil sampled for chemical analysis may be important for agrochemicals that undergo rapid transformations, especially if the soil concentrations are highly dependent on environmental conditions that can change abruptly within a small spatial area. The purpose of this study was to investigate the influence of six sample volumes on the magnitude and variability of several commonly measured surface soil parameters. This paper presents the results for the agrochemical and plant nutrient, nitrate-nitrogen. The smallest sample volume (slightly smaller than the most commonly used hand sampler) was often too small to accurately estimate the statistical parameters for soil nitrate-N. The smallest sample volume also required 20 to 50 percent more samples than the larger samplers to achieve satisfactory estimates of the sample mean. No clear trends in parameter estimation occurred across the range of larger sample volumes. This research provides better guidelines for the size of soil samplers that should be used to most clearly characterize the nitrate-N levels in a surface soil.

Technical Abstract:
The influence of sample volume on the magnitude and variability of soil NO3-N levels was investigated at Beltsville, MD, on a Beltsville silt loam soil (Typic Fragiudult). Five soil cores, ranging in volume from 38 to 366 cm3, and block samples of 8770 cm3 were collected from plow- and no-till plots before and after planting corn (Zea mays L.). Nearly all the NO3-N values were lognormally distributed. The magnitude of skewness was related more to NO3-N concentration than to sample size, except for the largest size (8770 cm3) which was always minimally skewed. The smallest sample volume was often too small to accurately estimate the statistical parameters for soil nitrate-N, and required 20 to 50 percent more samples than the larger sample volumes to achieve satisfactory estimates of the sample mean. No clear trends in parameter estimation occurred across the range of larger sample volumes.

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SOIL SCI. SOC. AM J.

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THE CALCULATION OF OPTIMUM NITROGEN FERTILIZATION RATES IN THE DEVELOPMENT OF NUTRIENT MANAGEMENT PROGRAMS

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WEISMILLER RICHARD A
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MEISINGER JOHN J

Interpretive Summary:
Throughout the Chesapeake Bay region nutrient management planning has formed the basis for more accurate forecasts of corn fertilizer N needs. The goal of these plans is to maintain a profitable agriculture while lowering environmental impacts. However, it must be realized that agriculture's environmental impact will never be entirely eliminated due to the complexities of the agriculture N cycle and uncertainties in rainfall. The N management plans are based on a careful N accounting of major N credits and debits. The N debit calculation is based on the realistic yield goal of the field. Nitrogen credits are then calculated for manure additions, based on manure analysis, and for previous legume crops. The final N accounting forecasts the need for supplemental fertilizer N. The final decision for fertilizer N can be checked with the pre-sidedress soil nitrate test which measures the nitrate concentration in the surface foot of soil when the corn is about 12 inches tall. This N soil test has successfully identified sites that are N sufficient and therefore can save over-application of fertilizer N. Future work will require further field testing and evaluation of these nutrient management systems.

Technical Abstract:
Improved methodology for the calculation of the optimal amounts of corn fertilizer N has formed the basis for Nutrient Management Planning throughout the Chesapeake Basin. The objective of these plans is to maintain a viable agriculture while lowering environmental impacts, although it must be recognized that owing to uncertainties in weather these impacts can never be entirely eliminated. The general form of the N management plan involves estimating the crop N requirement, the N credit for manure, and the N credit for crop rotations. The corn N requirement is estimated from the expected yield (in bu/a) multiplied by a factor of 1.0 lbs N/bu. Manure N credits are calculated from a manure analysis and actual application rates based on a calibrated manure spreader. Legume N credits are evaluated from the type of legume and the vigor of the stand. The adequacy of the forecasted N rate can be checked with the pre-sidedress N soil test which measures the nitrate-N content of the surface foot of soil when the corn is 12 inches tall. Future research in this area must include improvement and calibration of these predictive techniques and newer soil N tests. This continued research will require a commitment to applied field research.

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PROCEEDINGS OF CHESAPEAKE BAY RESEARCH CONFERENCE DEC 11 1990

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THE ANALYSIS OF 4-NITROPHENOL IN FOG WATER

VENKAT JYOTHSNA
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GLOTFELTY DWIGHT E

Interpretive Summary:
Pesticides may volatilize after they have been applied to crops. Airborne pesticides may be trapped in rain or fog droplets and returned to earth. Not only pesticides, but also their breakdown products are transported in the atmosphere. We have detected some of the breakdown products of parathion in fog water. One of these, 4-nitrophenol, the subject of our research and we have developed a rapid analytical method for measuring trace quantities of 4-nitrophenol in collected fog water based on a procedure employing solid phase extraction.

Technical Abstract:
The atmosphere plays a key role in the transportation and deposition of pesticides. Fog droplets are responsible for the removal of amounts of pesticides from the atmosphere. Samples of fog water from California have been found to contain a number of pesticides including parathion. Transformation products of pesticides also occur. Among these, 4-nitrophenol and paraoxon probably arise by atmospheric oxidation of parathion. An analytical method, based on solid-phase extraction followed by gas chromatography, has been developed to measure concentrations of 4-nitrophenol at the parts per billion (ppb) level in fog water.

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CHEMOSPHERE

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ENZYMZTIC PRETREATMENT FOR EXTRACTION OF STARCH ENCAPSULATED PESTICIDES FROM SOILS

WIENHOLD BRIAN J
GISH TIMOTHY J

Interpretive Summary:
Contamination of groundwater by agricultural chemicals is a problem receiving increasing attention. Much of the recent research has been directed at developing ways of modifying pesticide behavior in the soil. One modification that is currently being field tested in several states involves trapping the pesticide in a starch capsule. Evaluation of the effectiveness of starch encapsulation on behavior modification requires that methods are available to measure the amount of pesticide remaining in the soil. Existing methods are not adequate for recovering all pesticides remaining within the starch matrix, hence pesticide residue is underestimated. The present paper reports on a new method for releasing the pesticide from the starch capsule where it can subsequently be measured using standard recovery procedures. The new procedure involves treating the sample with an enzyme and heating the sample for one hour prior to extracting the pesticide from the soil. This new method adds approximately one hour to the recovery procedure and doubles the recovery of the pesticide from soil samples. This new method will be useful in studying pesticide behavior in field experiments when the pesticide has been applied in the starch encapsulated form.

Technical Abstract:
Recently a great deal of research attention has been directed towards modifying the behavior of agricultural pesticides suspected as nonpoint sources of groundwater contamination. One modification involves starch-encapsulation of pesticides which allows controlled release of the herbicide from a biodegradable matrix. Unfortunately, traditional analytical methods for recovering the parent compound trapped in the starch matrix are inadequate for chemicals with slow rates of release. Poor recovery prevents accurate mass balance assessment of these chemicals. An enzymatic (amylase) pretreatment was developed that allows quantitative recovery of the parent compound fro9m soil samples receiving the pesticide in the starch-encapsulated formulation. The inexpensive method adds approximately one hour to the extraction procedure and a two-fold increase in percentage recovery of starch encapsulated atrazine (2-chloro-4-ethylamino-6-isopropylamino-s-triazine). The enzyme pretreatment is not necessary for quantitative recover of pesticides that have rapid rates of release such as alachlor (2-chloro-2',6'-diethyl-N-methoxymethyl acetanilide). The method will be useful for studying how starch-encapsulation modifies pesticide behavior in the soil environment.

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WEED SCIENCE

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EFFECT OF WATER POTENTIAL AND SOIL MICROBES ON RELEASE OF STARCH ENCAPSULATED ATRAZINE AND ALACHLOR

WIENHOLD BRIAN J GISH TIMOTHY J

Interpretive Summary:
Increased concern over the fate of agricultural chemicals has lead to the development of chemical formulations aimed at reducing the susceptibility of chemicals to leaching and volatilization. One such formulation involves encapsulating the chemical in starch. Starch encapsulation successfully controls the rate at which a chemical is released into the soil but little is known about the effect environmental factors have on the rate of release. The purpose of this study was to determine what effect water availablity and soil microbial activity have on rate of release of starch encapsulated atrazine and alachlor. Results indicate that as availability declines rate of release also declines. Soil microbial activity increases the rate of release by digesting the starch matrix. Alachlor is released more quickly than atrazine most likely because alachlor is much more soluble in water than is atrazine. The environmental implication of these results is that controlled release reduces the susceptiblity of chemicals to undersirable leaching and volatilization losses. The agronomic implication of these results is that complete release of starch encapsulated atrazine may not occur for several weeks under dry conditions. Hence, herbicidal activity may not be realized for several weeks after application and weed control may be reduced at early times.

Technical Abstract:
This study was initiated to improve our understanding of how water potential and soil microbial activity influence rate of release of starch encapsulated atrazine (6-chloro-N-ethyl-N'-(1-methylethyl)-1,3,5-triazine-2,4-diamine) and alachlor (2-chloro-N-(2,6-diethylphenyl)-N-(methoxymethyl) acetamide). Water potential, imposed using polyethlene glycol, significantly influenced swelling of the starch matrix and rate of release of both herbicides. At a water potential of 0 MPa, complete release required 21 days for atrazine and seven days for alchlor. As water potential declinded so did rate of release. At a water potential of -1.5 MPa than 50% of the encapsulated atrazine and less than 80% of the encapsulated alachlor had diffused out of the starch matrix after 28 dyas. Soil microbes increase the rate of release. After 21 days there was a two-fold increase in the percentage of atrazine released from starch capsules applied to nonsterile soils compared to capsules applied to sterile soils. These results suggest that starch encapsulation is effective in controlling the rate of release of the herbicides used in this study, potentially reducing the susceptibility of these compounds to volatilization and leaching losses. However, full herbicidial activity may not be realized for 1 to 3 weeks after application when these herbicides are applied as starch encapsulated formulations.

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AQUEOUS OZONOLYSIS OF S-TRIAZINES. I. DESCRIPTION OF ATRAZINE DEGRADATION PATHWAY AND PRODUCT IDENTIFICATION

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Interpretive Summary:
Effective disposal of pesticide waste and equipment rinsate is desired to prevent contamination of groundwater and farm wells. A two stage process under investigation involves treatment of the waste with ozone followed by microbial degradation to give carbon dioxide, nitrogen, water and salts. Atrazine, one of the highest use herbicides, was found to be somewhat resistant to degradation relative to other pesticides. This study examined the overall mechanism by which ozone reacts with atrazine and the reaction products were isolated and characterized. Results demonstrated that the chlorine of atrazine was not removed and that the ring structure remained intact. This basic information is needed to further develop and optimize the waste disposal process.

Technical Abstract:
The aqueous ozonation of atrazine (2-chloro-4-ethylamino-6-isopropylamino-s-triazine) at pH 6 afforded four primary products: 6-amino-2-chloro-4-isopropylamino-s-triazine, 6-amino-2-chloro-4-ethylamino-s-triazine, 4-acetamido-2-chloro-6-ethylamino-s-triazine and 4-acetamido-2-chloro-6-isopropylamino-s-triazine. These compounds were subsequently degraded to 2-chloro-4,6-diacetamido-s-triazine, 4-acetamido-6-amino-2-chloro-s-triazine and 2-chloro-4,6-diamino-s-triazine. The amino alkyl groups are the first site of attack and are either removed or converted to the acetamide but not to the aldehyde. The s-triazine ring remains intact and the chlorine is not removed. Studies also demonstrated that the alkyl group is far more reactive than the amide moiety, which in turn is oxidized more rapidly than the amino group.

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SECTION 4

BARC GROUNDWATER SCIENTISTS

ADDRESSES AND PHONES

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